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Works by the same Author.

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1906. Constable, London. 21/– net.

STUDIES
ON IMMUNISATION

AND THEIR APPLICATION TO THE

DIAGNOSIS AND TREATMENT OF BACTERIAL INFECTIONS

By

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The Physician of the Future will be an Immunisator

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Dedication.

TO

ÉLIE METCHNIKOFF AND PAUL EHRLICH

THIS ACCOUNT OF AN ENDEAVOUR TO WIN

FROM THE INTELLECTUAL SEED SOWN BY THEM

A HARVEST FOR MEDICINE

IS DEDICATED,

AS A TRIBUTE OF FRIENDSHIP AND ADMIRATION,

BY

THE AUTHOR.

PREFACE

HAVE brought together in this volume the series of studies on the rotective elements of the blood, on vaccine-therapy, and on therapeuic immunisation taken generally which I have published during the ast dozen years.

The three final papers have been revised. The rest are reprinted ere without change, except only in the respect that the detailed deription of the technique, which I am reserving for separate treatment, as been omitted.

In Part I, I have placed those papers which deal primarily with the rotective elements of the blood, and less directly with the problem f immunisation. These are arranged in three sub-sections entitled spectively *agglutinins*, *bactericidins*, and *opsonins*.

In Part II will be found, arranged in the order of publication, the apers which deal with the real subject-matter of the book, i.e. with the roblem of fighting bacterial infection by those defensive agencies hich the organism itself employs when it contends with microbic vasion.

The papers in this section of the book fall naturally under three periods.

In the papers of the *first* period the conception of vaccine-therapy iginates and takes shape.

Just as the antirabies inoculations of Pasteur—inoculations which e undertaken after the implantation of the rabies virus into the body -are affiliated to his earlier prophylactic inoculations, so also here *phyctic* inoculation, if I may term it so, came to birth from *prophylactic* oculation.

When once it had become clear in the course of an investigation to the effect of anti-typhoid inoculation upon the blood, that it would practicable to control the "negative phase" and to confer upon a tient the advantages of immunisation without risk or appreciable lay, the thought lay very near that it might prove possible to elicit, en after microbes had made good their entry into the body, an imunising response which would be therapeutically valuable.

In the papers of the *second* period further points emerge.

These emerge in connexion with research undertaken to account for e failure of vaccine-therapy in individual instances, and to account for e irregularities of the opsonic readings obtained in certain types of fections.

It is brought out in these papers that the antibacterial agencies, even

when present in the circulating blood, may fail to come into effecti application upon the microbes in the focus of infection.

The significance of this fact is already emphasized in Part I, in a pap dealing with the distribution of agglutinins in the infected organism.

It is further brought out that instead of the immunisator having t field left free for his operations, he has, in certain classes of cases, to reck with spontaneous auto-inoculations which seriously complicate his tas

Upon the account of these new developments follows naturally discussion of the means of dealing with these new difficulties. Th follows upon this, in view of the progressive encroachment of vacci therapy upon spheres hitherto assigned to other methods of treating ba terial infections, inquiry into the justification for these methods a in particular into the justification for the methods of serum thera surgical extirpation, and antiseptic applications.

The papers of the *third* period express the views which I hold to-da There are three of these papers.

In the *first* I have attempted an exhaustive enumeration and a criti survey of the current methods of combating bacterial disease. Followi upon this, I have endeavoured to give a reasoned exposition of the ma principles of therapeutic immunisation, assigning in the proposed thera peutic programme to the inoculation of vaccines, and to the determinati of protective agencies to the seat of infection, to each its rôle. I th endeavour to show that (with hardly an exception) whatever in eith time-honoured or new methods of treating bacterial diseases posses value, may quite well derive that value from the fact that it carr out some portion of the programme of therapeutic immunisation. I co plete the paper by pointing out how the achievements of vaccine-thera ought to be judged, and what bacteriological knowledge and labour is quired for the proper conduct of this method.

In the *second* of this series of papers, so far as has been practicab each new contention made in the foregoing expository paper is substa tiated by detailed evidence.

And in the *last* paper, after a critical review of the position tak up by those physicians who are confident that they need in immuni tion no guide except such as is furnished by the clinical symptoms pass to consider the rationale and the practical carrying out of all th department of therapeutic immunisation which concerns the conveya of the antibacterial agencies of the blood into the focus of infection

It has formed no part of my design in these three final papers eit to give a detailed account of achievements of vaccine-therapy in different bacterial infections, or to supply a manual for treatment.

I have been concerned only with the establishment of a therapeu principle.

And inasmuch as the success of that principle both in the case staphylococcus and tubercle infections is abundantly illustrated in earlier papers, and inasmuch as successes quite similar to those just

rred to are achieved every day in connexion with innumerable other fections, I submit that the principle of therapeutic immunisation, like e principle of prophylactic inoculation, is a principle of general applition.

I go even further; I submit that the principle of *phylactic* inoculation—at is to say, the principle of building up the resisting power of the stem against any microbe which may have entered the body—will timately hold its own even against the principle of warding off infection om the susceptible patient.

It is to me conceivable that curative medicine may give us even ore effectual aid against bacterial diseases than has either hygiene and eptic surgery.

Passing from the subject matter of the book to the method in which at subject matter is presented, I am conscious that something in the ture of an apology or explanation is required, for the fact that the position is in the form of a succession of separate papers.

As compared with an exposition in which every point is set out *initio* in an orderly manner, and is placed at once in the proper erspective, a collection of papers such as is here offered lays itself open criticism on the score of frequent repetitions and of the fact that the rlier presentations of the theme are subject to correction by the later positions.

I would urge that the method which is here adopted may none the ss have compensating advantages.

We may take it that no one ever really grasps a new body of facts y following any one exposition—even the most skilful exposition. In ct, the more skilful, i.e. the more fluent and unimpeded, the exposion, the less deeply do the new facts penetrate into the mind, and the ss permanent is the impression. This is so because the reader shown only one aspect of the facts, and because these are shown to m, not from the point of outlook which he himself would have sected, but from some unfamiliar outlook to which, in consideration of e exigencies of the exposition, he has suffered himself at the outset be transported.

In every advance to new knowledge, it is above all essential that the d experience should be linked up with the new. We ought to start om what is familiar to us, ought to choose our own paths of approach, ught ever and again to retrace our steps to make sure of our bearings, d finally, on arriving upon the new ground, we ought to find at nd an efficient guide.

That this and no other is the royal road to knowledge will be borne upon us if we reflect that all our effective knowledge—except, of urse, such as we may have elaborated for ourselves—is knowledge hich has come to us by personal converse, by question and answer, d by demonstration; and the same will come home to us if we recall w urgent is the impulse which impels us, when an author or lecturer

has broken new ground, to seek from him an answer to this and that, so that we may link up at these points our experience with his.

Now in the case like the present, where an unfamiliar doctrine a an unfamiliar body of facts is set forth, some of the difficulties wh stand in the way of its effective apprehension may, if I mistake not, removed by supplying to the reader, in place of a single consecut exposition, a series of expositions such as are here furnished. That, any rate, has been my thought.

There remains the difficulty that no collection of separate pape such as is here presented, can possess such an organic unity in its structu design as would enable the reader who is referring to the book to place finger upon the place where the particular point he is in search of mig properly be found.

With a view to remedy this defect I have elaborated the ind making it, I would hope, something in the nature of an orde synopsis.

Incidentally, I have been able to turn the index to account a in other ways.

I have, by a certain exercise of selection in the matter of the entri found it possible to give prominence in the index to certain points whi by reason of the brevity with which they are treated in the text, mig quite well fail to make good their claim to the thoughtful considerati of the reader.

I have also found in the index an agency through which repl might be furnished to certain of my critics. By making use of t index as a finger-post, I have pointed these critics in each case to impersonal reply inserted in the text. It is a method of rejoinder wh I would venture, with all submission, to recommend.

It remains to me, in bringing my task to a close, to express my friend Dr. William Bulloch and to his pupils, Drs. Atkin, Weste and Keith, my grateful acknowledgments for their permission reprint their papers among my own. To my friends, Col. C. Bi R.A.M.C., Major George Lamb, I.M.S., Major F. Smith, R.A.M. Major F. N. Windsor, I.M.S., and Staff-Surgeon S. T. Reid, R.I who have worked with me in the past, and to Capt. Stewart R. Dou las, I.M.S, (retired), Dr. J. Freeman, Dr. J. H. Wells, and Dr. Alexan Fleming, who are working with me now, I desire to express simi acknowledgments with respect to our joint work.

I have also to thank the Proprietors of the *Lancet*, *British Medi Journal*, *Clinical Journal*, and *Practitioner*, and the Council of the *Ro Society* for permission to reprint papers originally published in th Journals.

I have further a debt of special obligation to discharge to my frie Lord Justice Fletcher Moulton for continuous help both in the form criticism and suggestion in the preparation of this book.

My grateful thanks are also due to Mr. A. Bazire for assistance in preparation of the index.

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THE ANTIBACTERIAL ELEMENTS OF THE BLOOD FLUIDS

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Part I

ON THE ANTI-BACTERIAL ELEMENTS OF THE BLOOD FLUIDS

On the
Application of the Serum Test
To the
ferential Diagnosis of Typhoid and Malta Fever
And on the
Application of the Method of Serum
gnosis to the Elucidation of certain Problems in onnexion with the Duration of Immunity and the Geographical Distribution of Disease.[1]

By A. E. Wright and Surgeon-Captain F. Smith, A.M.S.

om the Laboratory of the Pathological Department, Army Medical School, Netley.

notorious that every clinician in tropical and sub-tropical countries stantly meeting cases of continued fever of which he finds it impos- to say whether they are cases of typhoid fever, of Malta fever, of rial fever, or of some other (possibly as yet undescribed) variety of nued fever. In cases of this kind, and further in cases where it is ble that a diagnosis should be arrived at early in the course of the se, the prompt and accurate methods of diagnosis which medical rch has recently placed at our disposal will be everywhere welcomed. ree of such methods of diagnosis are at present available.

he *first* place, the blood may be examined microscopically with a view termining the presence or absence of pathogenic micro-organisms. method is of particular utility in connexion with the determination e presence or absence of the micro-organisms of malaria and of um fever.

the *second* place, the excretions (and, in particular, the urine [2]) may

eprinted from the *Lancet*, March 6, 1897.

he importance both from a diagnostic and a hygienic point of view of recog- the presence of typhoid bacilli in the urine was pointed out in a paper which itten by one of us in conjunction with Surgeon-Major Semple (*The Lancet*, , 1895). The results which were arrived at in that paper have been recently ied and somewhat extended by Dr. P. Horton Smith (Royal Medical and gical Society, February 7, 1897).

be examined by appropriate cultivation methods with a view determining the presence or absence of pathogenic micro-organi This method is applicable to typhoid fever. It is probably applicable to Malta fever.

Lastly, the method of serum diagnosis may be applied. By this me the blood of a patient who is suffering, or who has suffered, from a co ued fever is tested in turn with any pathogenic micro-organisms may be available, and in particular with the micro-organisms which causally associated with typhoid fever and Malta fever. We recently been applying this serum test to the diagnosis, and espec to the after-diagnosis, of tropical and sub-tropical continued fe Convalescents from these fevers come under observation in Netley in persons of soldiers and others who are invalided home from mili stations abroad.

Examples of the Application of the Method.

The following are typical examples of the application of the me of serum diagnosis to the differentiation of Malta fever from typ fever.

Example 1.—Sergeant L——, of the Medical Staff Corps, arr from Aldershot in October, 1896, having been detailed for duty at Ne A few days after arrival at Netley the patient "reported sick," and admitted to hospital with a recrudescence of a fever from which he suffered some months before. His papers showed that this fever had diagnosed to be typhoid fever. The patient's serum showed absolu no reaction with the bacillus typhosus. His serum in 10,- 25-, 50-, and 200-fold dilutions showed a very marked reaction with the micro cus Melitensis of Bruce. Inquiry into the patient's history revealed when he went into hospital at Aldershot he had only recently retu from Gibraltar.

Example 2.—Sergeant F——, of the Medical Staff Corps. patient was admitted to hospital in Malta on October 2, 1896. He in hospital on November 20. Some of his symptoms, such as diarr associated with blood in the stools, were suggestive of typhoid fe The necropsy, however, showed only slight ulceration of the colon, the absence of any implication of Peyer's patches. In the light o facts, his case was diagnosed as having in all probability been a ca Malta fever.

A stepson of the above, aged nine years, contracted fever in M and died at the end of November after ten days' illness. The case diagnosed as probably one of typhoid fever.

Another stepson, aged seven and a half years, contracted feve Malta at the beginning of last December, and was immediately inva home. Doubts were expressed in his papers as to whether the case one of Malta fever or of typhoid fever. He is now quite convales

serum gives an absolutely negative reaction with the micrococcus
ensis, but gives a very marked effect in 10-, 25-, 50-, and 100-fold
ions with the bacillus typhosus.

he wife of Sergeant F—— contracted fever in Malta in the month
ptember. She was in bed for ten days. The serum gives an abso-
y negative reaction with the bacillus typhosus. The serum, however,
in 10-, 25-, and 50-fold—but not in 100-fold—dilutions a character-
reaction with the micrococcus Melitensis.

child of Sergeant F——, a girl aged three years, contracted fever in
a in May, 1896. She suffered severely from fever till the end of
ember, 1896. She is now quite convalescent. Her serum gives
utely no reaction with the bacillus typhosus, but the serum in 10-,
50-, 100-, and 200-fold dilutions gives very marked reaction with the
ococcus Melitensis.

t will be obvious how very difficult a clinical problem was presented
nis last series of cases, and how very easily clinical problems of this
can be re-solved by the employment of the method of serum
nosis.

xample 3.—Private P—— (at present under treatment in Netley),
ided home from India for secondary syphilis. He has suffered for
ast four months from a continuous but very irregular fever, which
ome resemblance to the typical curve of Malta fever. Examination
story discloses that the patient left England in March, 1892, for Gib-
r. He continued in good health until the autumn of that year,
he was attacked with continued fever, which, from its long duration
days in hospital) and from the fact that the patient was constipated
ighout, may not unreasonably be assumed to have been Mediter-
an fever. The patient left Gibraltar for India with his regiment in
mber, 1893, feeling perfectly well. In India he was noted as having
red occasionally from attacks of fever. These attacks of fever were,
ever, not severe enough to require treatment in hospital. The
nt began to suffer again from fever very soon after he was put under
rs for home. His blood has been examined by us with negative
ts for malaria, and his urine has recently been examined by us with
tive results both for the presence of typhoid and Malta fever micro-
nisms. His serum exerts no influence upon the bacillus typhosus.
anifests, even in 1,000-fold dilution, a very characteristic effect upon
nicrococcus of Malta fever. It is obvious that the interpretation of
esults of serum-diagnosis was here complicated by the fact that the
nt has presumably suffered some four years ago from Malta fever
that a reaction with the micrococcus of Malta fever might possibly
persisted for this period. In view, however, of the fact that the
n shows the specific reaction in such an enormous dilution, and,
er, in view of the fact that four years have elapsed since he suffered
fever in Gibraltar, and lastly, in view of the fact that the patient

is now suffering from pains in the joints, it seems to us practically cert
that this person is suffering from Malta fever.[1]

Having sufficiently illustrated this last point, we may now subjoi
tabular form the results which we have obtained by the applicatio
this method of serum diagnosis.

*The results which are tabulated below were obtained by testing the reac
of the serum in the capillary sedimentation pipettes.*

*In the tables which are subjoined a voluminous sedimentation is indic
by the sign* + ; *a slighter but still perfectly characteristic sedimenta
is indicated by the sign* * ; *a negative result is indicated by the sign* 0.

Table I.—*Cases diagnosed as "Malta Fever," which showed Reac
on the Micrococcus Melitensis of Bruce.*[2]

No. of Case.	Period elapsed since Illness.		Degree in which Serum was Diluted.				
	Years.	Months.	1 in 10.	1 in 25.	1 in 50.	1 in 100.	1 in 2
1[3]	—	5	*	*	*	0	0
2[4]	—	6	+	+	+	—	—
3	—	6	+	+	+	+	+
4	—	8	+	+	+	+	+
5	1	8	+	+	+	+	+
6[5]	1	9	+	+	+	+	+
7	3	—	+	*	*	0	+
8[6]	Now ill.		+	+	+	+	0

[1] The question of greatest clinical interest in connexion with this case is
course, the question whether the patient was re-infected with Malta fever in In
or whether the attacks of fever from which he suffered at intervals in India v
mere recrudescences of his original Malta fever. In view of the facts which on
us has, in conjunction with Surgeon-Major Semple, elicited on the survival of
micrococcus Melitensis in the spleen of monkeys who have perfectly recovered f
Malta fever, both these interpretations appear to us to be equally admissible.

[2] We are deeply indebted to Surgeon-Captain L. Hughes for sending us a cul
of this micro-organism. The culture which was sent to us was obtained in M
from a fatal case of the disease.

[3] Said to have been a very slight attack.

[4] Higher dilutions not tried.

[5] Marked effect in this case up to 1 in 300.

[6] Gives a positive reaction up to, but not above, a 1,000-fold dilution.

…LE II.—*Cases diagnosed "Typhoid Fever" which showed Reaction on Typhoid Bacillus.*

. of se.	Period elapsed since Illness.		Degree in which the Serum was Diluted.				
	Years.	Months.	1 in 10.	1 in 25.	1 in 50.	1 in 100.	1 in 200.
	—	5	+	+	*	0	0
	—	6	*	*	0	0	0
	—	6	+	+	+	+	+
[1]	—	6	+	+	+	—	—
	—	6	*	*	0	0	0
	—	7	*	*	0	0	0
[1]	—	9	+	+	+	—	—
	—	9	+	*	0	0	0
	—	10	+	*	0	0	0
	1	2	*	*	0	0	0
	1	8	+	+	*	0	0
	2	—	+	*	0	0	0
[1]	6	—	+	+	—	—	—
	6	—	+	*	0	0	0
	12	—	*	*	0	0	0

…LE III.—*Cases diagnosed "Typhoid Fever" which had no Effect on the Typhoid Bacillus, but showed a distinct Reaction on the Malta Fever Micrococcus.*

of e.	Place in which Illness Occurred.	Period elapsed since Illness.		Degree to which the Serum was Diluted.				
		Years.	Months.	1 in 10.	1 in 25.	1 in 50.	1 in 100.	1 in 200.
	Aldershot . . .	—	9	+	+	+	+	+
	Sabathu, India[2] .	—	7	+	+	+	+	+
	Sabathu, India[2] .	—	7	+	—	—	—	—

…LE IV.—*Cases diagnosed "Typhoid Fever" which showed no Reaction on either the Typhoid or Malta Fever Organism.*

…o. of Case.	Place in which Illness occurred.	Period elapsed since Illness.	
		Years.	Months.
2, 3, and 4	Sabathu, India	—	8, 8, 6, and 6
5	Nowshera, India	—	7
6	Lucknow, India	—	10
7	Line of march, India . . .	—	3
8	Rangoon, Burmah	—	6
9	Shwebo, Burmah	—	9
10	Cannanore, India	—	10
11	Hong-Kong	—	9
12	Benares, India	—	8
13	Umballa, India	—	6
14	Natal, South Africa . . .	17	—

[1] Higher dilutions not tried.
[2] India was the only foreign station in which the patient had served.
[3] Higher dilutions not tried.

TABLE V.—*Cases diagnosed "Malarial Fever," or not specifically diagnos[e]* *which showed Reaction on the Malta Fever Organism.*

No. of Case.	Place in which Illness occurred.	Period elapsed since Illness.		Sedimenting Effect.				
		Years.	Months.	1 in 10.	1 in 25.	1 in 50.	1 in 100.	1 in [illegible]
1[1]	Nowshera, India .	—	11	+	+	+	+	+
2	Hong-Kong . .	—	6	*	*	0	0	0
3[2]	Meean-Meer . .	Now ill.		+	+	+	+	+

TABLE VI.—*Case diagnosed "Malta Fever" which showed no Reaction* *the Micrococcus Melitensis, but which showed a Distinct Effect on t* *Typhoid Bacillus.*

No. of Case.	Place in which Illness occurred.	Period elapsed.		Degree to which the Serum was Diluted.				
		Years.	Months.	1 in 10.	1 in 25.	1 in 50.	1 in 100.	1 in 20
1	Malta	Ill at time of examination		+	+	+	*	0

A not inconsiderable number of interesting facts are disclosed by study of these tables. The following are perhaps the more important these :—(1) Consideration of Table I discloses the fact that the micr coccus Melitensis, which was discovered by Surgeon-Major D. Bruce, in reality, as he asserted it to be, the true cause of Malta fever. Out nine clinically more or less well-characterized cases which were examin by us, the eight which are tabulated here have shown a perfectly characte istic reaction to the micrococcus Melitensis. The only one of the ni cases which failed to react was that of a patient who had suffered fro Malta fever five years previously to the date of our examination. We ma not unreasonably surmise that the specific power of sedimenting the micr organisms of Malta fever may, in this case, have passed away. (Consideration of Tables III and V discloses the fact that Malta fever as Bruce and others have long surmised, a disease which is not by a means confined to the Mediterranean basin. We have here what take to be definite evidence of its existence in three stations in Northe India. We have, further, what we take to be probable evidence of existence in Hong-Kong. (3) Table III shows that a certain numb of cases which are from their clinical symptoms diagnosed to be typho fever, are in reality cases of Malta fever. (4) Table IV shows that considerable number of cases which are from their clinical characte diagnosed to be typhoid are in all probability neither cases of typho fever nor cases of Malta fever. It is possible that the fevers from whi

[1] India was the only foreign station in which the patient had served.
[2] Distinct effect up to 1,000-fold dilution.

se particular patients suffered may have been malarious in character. s also possible that these fevers may have belonged to some as yet lefined category of continued fever. What is important for us to e is the fact that any tropical fever which is unaccompanied by the sence of malaria parasites in the blood, and which can by the thod of serum-diagnosis be shown to be neither typhoid fever nor lta fever, is a fever which might very profitably be made the subject bacteriological investigation. (5) Tables I and II show that the cific agglomerating and sedimenting power, whch is acquired by blood in cases of fever, persists in the blood for a considerable n of years. We have been able to demonstrate its existence in the od in the case of typhoid fever after no less a period than twelve rs. Similarly, we have been able to demonstrate its persistence in blood for at least three years after an attack of Malta fever.

These facts are of the greatest importance, if, as was urged in a er on typhoid vaccination [1] which was written by one of us in con- ction with Surgeon-Major D. Semple, the sedimenting and agglomera- g power of the blood is in reality a true index of the condition of nunity.

[1] Vide *British Medical Journal*, January 30, 1897.

A Note on the Occurrence of Malta Fever in India.[1]

By A. E. Wright and Surgeon-Captain F. Smith, A.M.S.

From the Laboratory of the Pathological Department, Army Medica School, Netley.

We have elsewhere [2] in a paper on the application of the method c serum diagnosis to the differential diagnosis of typhoid and Malt fever directed attention to the fact that Malta fever probably prevail not only in the Mediterranean basin, but also in India. We have sinc the date of the publication referred to further investigated this questior and we have found confirmation of our conclusions.

In view of the importance of the question, and in view further of th fact that public attention has recently been called (in Parliament an elsewhere) to the extreme prevalence of typhoid fever in an Indian statio (Sabathu), where, as our serum examinations teach us, Malta fever mus be very prevalent, we desire to be allowed briefly to refer to the matter

Our observations have been made on soldiers who have recently bee invalided home from India to the Royal Victoria Hospital, Netley. A minute quantity of blood was drawn off from the finger of each of thes invalids. This blood was diluted and was then examined in capillar sedimentation tubes in the manner which was recently described by on of us in the *Journal.*[3]

The results of our examinations are subjoined below in tabular form

In confirmation of the results which we have obtained by the appli cation of the serum test to the after-diagnosis of these cases of continue fever, we may mention that many of these patients have since thei arrival in Netley suffered from the ordinary sequelæ of Malta fever (suc as swollen testicle, sciatica, and rheumatoid joint pains), and that other have suffered from definite relapses of fever. Further, we may direc attention to the fact that careful Indian observers have described case of "atypical typhoid" which presented a set of symptoms and a temper ature curve which we now know to be almost characteristic of Malt fever. In this connexion we would particularly direct attention to a able report on typhoid fever in India which was published by Brigade Surgeon Marston in the Appendices to the Army Medical Report for 187

[1] Reprinted from the *British Medical Journal*, April 10, 1897.

[2] *Lancet*, March 6, 1897 (*vide* p. 3.) [3] *British Medical Journal*, January 16, 189

Table of Cases examined.

No. of Case.	Station from which invalided.	Regiment.	Whether Served previously in the Mediterranean.	Whether had suffered from Fever while on Service in Mediterranean.	Highest Dilution in which Sero-sedimentation Effect was obtained upon the Micrococcus Melitensis of Bruce.	Remarks.
1	Sabathu . .	Black Watch	No	—	1 in 150	Higher dilutions not tried in this case.
2	" . .	" "	No	—	1 in 10	
3	Nowshera . .	Argyll and Sutherland Highlanders	No	—	1 in 200	
4	Meean-Meer .	Somerset Light Infantry	Yes, in 1893	Yes	1 in 1,000	All these served in Sabathu before going to Meean-Meer.
5	" .	" " "	"	No	1 in 200	
6	" .	" " "	No	—	1 in 1,000	
7	" .	" " "	Yes, in 1893	No	1 in 200	
8	" .	" " "	"	No	1 in 300	
9	" .	" " "	"	No	1 in 150	
10[1]	" .	" " "	Yes, in 1892	No	1 in 150	

[1] This patient had enteric fever in Madras before he went to Sabathu.

Mediterranean or Malta Fever

With Special Reference to the Specific Agglutinating Substances which make their Appearance in the Blood in the course of that Disease.[1]

By Major C. Birt, R.A.M.C., and Captain G. Lamb, M.B. (I.M.S.).

From the Laboratory of the Pathological Department of the Army Medical School, Netley.

History of the Bacteriology of Malta Fever—Biological Characteristics of the Micrococcus Melitensis—Causal Association of the Micrococcus Melitensis with Malta Fever—The Effect of Specific Serum on Micrococcus Melitensis—Data Bearing on the Early Appearance of Agglutinins in the Blood—The Persistence of the Agglutinins in the Blood after Recovery—Incubation Period for Man—Geographical Distribution—Data Bearing on the Evolution of the Agglutinins during the Course of Malta Fever—Inferences which may be drawn from the above Observations—Value of an Estimation of the Agglutinating Substances as an Aid to Prognosis

Mediterranean fever is a disease too seldom recognized. It is often disguised under such names as simple continued fever, sweating typhoid,[2] etc., accounts of which still unfortunately find a place in current medical literature. Clinically the main features of Malta fever are briefly stated as follows: pyrexia of a more or less chronic type, accompanied by constipation and copious perspiration. Relapses are frequent, and convalescence is often retarded by neuralgias, joint-pains, and orchitis. This fever is the cause of much sickness among the British troops stationed at Malta and Gibraltar. Many men in the convalescent stage are invalided to England, and come under treatment in the Royal Victoria Hospital, Netley. Among these men relapses are not unfrequent after their arrival, and thus a wide field for study of the disease is afforded.

History of the Bacteriology of Malta Fever.

In 1887 Bruce[3] isolated a micro-organism in pure culture from the spleens of nine fatal cases and from blood drawn from the spleen during life in two instances. This organism he named the micrococcus Melitensis.

[1] Reprinted from the *Lancet*, Sept. 9, 1899.
[2] Jaccored: *Journal de Médecine*, March 10, 1899.
[3] *Practitioner*, September, 1887, and April, 1888; *Annales de l'Institut Pasteur* 1893, vol. vii, p. 289.

e succeeded in infecting monkeys by subcutaneous injection of small ıantities of the pure culture of this coccus. In these animals the micro-ccus Melitensis gave rise to a continuous fever, not unlike that in man, ıd usually caused death after a variable time. In the monkeys which ed from this infection Bruce found in pure culture in the spleen the me bacterium as he had inoculated. Hughes [1] has corroborated these servations of Bruce. He has isolated the micrococcus Melitensis from e spleens of fourteen men who succumbed to this disease. Further, he ıs infected monkeys with the cultures of the organism and, like Bruce, terwards recovered the micro-organism from the spleens of the animals ıich died. Gipps [2] has also obtained the same micrococcus in two fatal ses. In this country we have grown the microbe from the blood and leens of the only two fatal cases which have come under our notice.[3] ıe observations of the disease in infected monkeys, as described by ruce and Hughes, have been amply confirmed in this laboratory by a ng series of experiments undertaken some two years ago by Professor right and Major D. Semple with an organism sent from Malta by Hughes. e also have infected monkeys with the cultures obtained from the two tal cases mentioned above. As the details of these experiments coin-de in almost every particular with the descriptions previously given by ruce it is unnecessary to enter further into them. Durham [4] has shown at the infection can be conveyed to rabbits and guinea-pigs (animals garded by Bruce as immune to the disease) by means of intra-cerebral ıd intra-peritoneal inoculation. These observations have also been nfirmed in this laboratory by Professor Wright and Major D. Semple. arious attempts have been made by us to isolate the organism from the ine during the course of the fever in man. These have in all cases been ısuccessful. The plates either remained sterile or were overgrown by ore quickly growing bacteria. No bacteriologist has described the icrococcus Melitensis as having been isolated in any other disease.

Biological Characteristics of the Micrococcus Melitensis.

These have been fully described by Bruce,[5] Hughes [6] and Durham.[7] A ief *résumé* of their and our own observations will therefore suffice. The icrococcus Melitensis is a small coccus or cocco-bacillus about 0·33μ diameter, the bacillary form being more pronounced when grown on latin. We have not been able to satisfy ourselves as to the presence any motility other than Brownian movement. Gordon,[8] however, gures it as possessing usually a single and sometimes three or four short agella. Neither Durham [9] nor the workers in this laboratory have been

[1] *Mediterranean, Malta, or Undulant Fever*, London, 1897.
[2] *Transactions of the Epidemiological Society of London*, vol. ix, p. 76.
[3] These cultures had the same biological characters as the cultures supplied Hughes to this laboratory.
[4] *Journal of Pathology and Bacteriology*, December, 1898.
[5] Loc. cit. [6] Loc. cit. [7] Loc. cit.
[8] *The Lancet*, March 11, 1899, p. 688. [9] Loc. cit.

able to confirm this observation. It stains readily with all the basic aniline dyes, while it does not retain its stain by the Gram-Weigert method. The growth of this microbe on all media is characterized by its slowness. On agar (at 37° C.) no colonies are visible to the naked eye for at least two days. In cultures made directly from the spleen or blood in fatal cases the agar appears sterile to the unaided eye for from four to six days. The colonies, when developed, are small, transparent and dew-like, and are often limited to the lower end of the tube. Broth (at 37° C.) becomes turbid, the turbidity appearing on the second or third day. After a time there is a deposit of flocculi, the broth still, however, remaining turbid. Short chains are also found in this medium. If the specific serum is added to the broth the fluid remains clear, and a compact, felt-like growth takes place at the bottom of the tube in the course of weeks. On gelatin the growth is extremely scanty and slow; there is no liquefaction. It may be further noted that on media which are highly alkaline there is little or no growth. Its slow growth, the appearance on agar, and the microscopical characters will generally lead to its identification. The behaviour of the culture in question in the presence of the specific serum will confirm with certainty the diagnosis. In bacteriological literature no description of another microbe possessing the same biological characters can be found.

Causal Association of the Micrococcus Melitensis with Malta Fever.

The evidence collated above shows (*a*) that the micrococcus Melitensis has been recovered from almost every case of Mediterranean fever which has been examined bacteriologically after death, and has not been found elsewhere; (*b*) that the micrococcus Melitensis is capable of infecting monkeys, giving rise in them to a continuous fever not unlike that in man, and usually causing death, and that by special methods rabbits and guinea-pigs have also been infected; and (*c*) that from all these animals after death the same organism has been recovered in pure culture from the blood and organs, and after cultivation this has again occasioned the disease in other animals. The final link in the chain—i.e., the proof that the micrococcus Melitensis can produce Malta fever in man—has not heretofore been forthcoming. This is now furnished by two cases which have occurred as a result of inoculation with the micrococcus Melitensis; and by a third case the method of infection in which is not quite so definite; all these cases happened among the staff of this laboratory. They are given here in chronological order.

CASE 1.—On September 17, 1897, D.S. accidentally scratched himself with the needle of a syringe with which he had just injected into a horse a living growth of micrococcus Melitensis.[1] He immediately sucked the minute wound, plunged the hand into a 5 per cent. solution of phenol

[1] This culture was derived from a culture which had been isolated two years previously by Hughes from the spleen of a fatal case of the disease in Malta.

ch he had by his side, and almost at once cauterized the puncture with e phenol. But all to no purpose, for on October 2, fifteen days later, temperature rose and he went through a typical attack of Malta er (*vide* Chart 1).

CASE 2.—On March 1, 1898, A.E.W., in connexion with some experiments arding the elaboration of a method of vaccination against Malta fever, cted into his arm $\frac{1}{100}$th of an agar tube of a seven-day growth of rococcus Melitensis.[1] On March 17, sixteen days later, febrile symptoms in and pursued a course characteristic of this fever (*vide* Chart 2). s worthy of note that, as in most bacterial infections, there was a ked increase in the number of polynuclear white cells in the blood before the onset of fever.

CASE 3.—In February, 1899, Corporal S., the head attendant of the oratory, who was constantly employed in bacteriological operations this microbe, contracted an illness which soon assumed the typical racters of Malta fever and which was complicated by severe double itis (*vide* Chart 5). The method of infection could not be definitely ed in this case.

We shall have occasion to refer in more detail to these three cases in nexion with the question of the estimation of the agglutinating subces. We may here add that the diagnosis was confirmed in all three s by the serum sedimentation reaction. The reaction took place in dilutions of the serum, and during the course of the fever there were siderable variations in the agglutinating power of the serum. In the two cases the agglutinating reaction still persists a year and a half a year respectively after convalescence.

The Effect of Specific Serum on Micrococcus Melitensis.

If the above observations left any room for doubt as to the causal tion of this organism with Malta fever, that doubt would be comely removed by the investigation through which Professor Wright blished that the serum of patients who are suffering from or who have vered from Malta fever produces a specific agglutinating reaction on micrococcus Melitensis. This observer[2] has published fourteen obserons in which this coccus was sedimented after agglutination by high tions of the sera of patients who were suffering from or had recently vered from Mediterranean fever, and he showed that the agglutina- and sedimentation which were obtained in these cases with the us was more marked and definite than the corresponding reaction ch is obtained with the bacillus typhosus in typhoid fever. In a e recent publication, along with Semple,[3] he has further shown that l cultures of the bacteria can be employed with the same result as g cultures. During the course of our investigations we have arrived

[1] This culture was derived from a culture which had been isolated two years iously by Hughes from the spleen of a fatal case of the disease in Malta.

[2] *The Lancet*, March 6, 1897, p. 656 (*vide supra*, pp. 3–9.)

[3] *Brit. Med. Jour.*, May 15, 1897.

at a like conclusion. These observations have been confirmed by oth observers. Aldridge[1] has reported fourteen cases in Malta in which tl reaction was obtained, and Elkington[2] has detected agglutination of tl microbe seventy-five times in 158 samples of blood of various febr patients in Gibraltar. Durham[3] has likewise obtained this phenomen with the blood of rabbits and guinea-pigs which he had infected by mea of intra-cerebral and intra-peritoneal inoculation. Further, the test being daily employed in the laboratory of the Public Health Departme of Malta.

Since the date of Professor Wright's original publications we ha carefully investigated this reaction. It is the result of this investigati which we have now to present and which we shall preface by giving short account of the methods employed. All observations were ma microscopically by means of the sedimentation tubes (rather less th one millimetre in diameter) devised by Professor Wright.[4] Equ quantities of the serum, diluted with normal salt solution, and of ster emulsions of the micrococcus were employed in all cases. T emulsions were prepared from fresh agar cultures with normal sali solution.

It is obvious that the sedimentation reaction of any serum wh one and the same culture is used, will vary with the number of bacte which the emulsion contains. In order to make the quantitative estim tions which are here reported comparable amongst themselves the follo ing procedure was adopted. (1) The emulsions were all prepared in exac the same way. A uniform growth in agar of from five to seven da was obtained. This was emulsified with sterile normal saline solutio a fixed quantity of which—viz., 0·25 cubic centimetre—was used for eve square centimetre of agar culture. The bacteria were then killed by heati at 60° C. for from ten to fifteen minutes and finally 0·5 per cent. ca bolic acid was added. In this way emulsions containing approximat the same number of bacteria were obtained. (2) When the estimati of the agglutinins was made during the whole course of a case the sa emulsion, if possible, was used throughout. If a fresh emulsion had be substituted the sedimentation reaction of the same specimen of ser was compared in the two emulsions and any difference taken into accou in recording the results. Following this method we first examined controls more than 50 samples of blood of healthy people, among wh were eleven negroes, and 101 samples taken from patients suffering fr the following diseases: enteric fever, 38 cases; malarial fevers, cases; liver abscess, 4 cases; local suppurations, 10 cases; ac streptococcic infections, 5 cases; rheumatic fever, 5 cases; tub

[1] *The Lancet*, May 21, 1898, p. 1394: Report of the Health Departmen Malta, 1898.

[2] *Reports of the Sanitary Officer of Gibraltar*, 1897 and 1898.

[3] Loc. cit.

[4] *British Medical Journal*, February 5, 1898.

osis, 5 cases; secondary syphilis, 6 cases; cancer, 2 cases; dysentery, uses; acute tonsilitis, 1 case; and diabetes, 1 case. In the course of se estimations we found that nearly all the specimens of blood examined e a well-marked, sometimes complete, sedimentation in dilution of 1 2. Many gave a faint trace in dilution of 1 in 10, but the majority not give any reaction in that dilution. In none of the above instances we ever observe a complete sedimentation to occur in a 10-fold dilu-, nor did we ever see a trace of sedimentation in a 20-fold dilution. ring thus fixed the limit of the agglutinating power of normal sera l of sera derived from patients suffering from other diseases, we may ceed to consider the cases in which sedimentation above these normal its was observed. We have notes of over 120 individuals in whom this ction was found, and as many cases were examined on several occasions have made several hundred observations in all. A list of these cases h dilutions in which the serum reacted is given in Table I and Table II.

On referring to the above tables it will be seen that fifty-three cases came ler observation while still suffering from Malta fever, the remainder rarying periods after convalescence had been established. In forty-four rile cases in which the sedimentation value was quantitatively deter-ned the average dilution for a complete reaction [1] was between 600 and , though the limits were very wide; one serum was tested in which nplete sedimentation took place in a 6,000-fold dilution. When we trast these figures with those obtained by us in cases of typhoid fever, find that as compared with the serum of typhoid patients the serum Malta fever patients sediments its homologous micro-organism in con-erably higher dilutions. Moreover, the clumps of agglutinated micro-ci which are deposited are much more compact and are better defined n those of Eberth's bacillus. Indeed, the serum reaction of Malta er is one of the most delicate bacteriological tests with which we are uainted, and ought undoubtedly to supersede the ordinary clinical thods of diagnosis, especially as it occurs, as we shall show later, com-atively early in the course of the disease. By its means we have recog-ed cases invalided home as gonorrhoeal rheumatism, simple continued er, debility, rheumatic fever, enteric fever, malaria, etc.

ata bearing on the Early Appearance of the Agglutinins in the Blood.

In experiments on monkeys performed by us, and in those pre-usly referred to as done by Wright and Semple, it is definitely wn that the agglutinating substances may appear in the blood early as the fifth day after inoculation of the living virus, and t they are well developed by the ninth or eleventh day. The ves of the sedimentary values of the sera of a series of guinea-pigs

[1] In these cases traces of sedimentation occurred on an average up to 1,000-dilution.

TABLE I.—*Cases of Malta Fever examined during the Course of Disease.*

No. of Case.	—	Dilutions of Serum.																						Remarks.
		10	20	30	50	70	80	100	150	200	300	400	500	600	800	1,000	1,200	1,500	2,000	4,000	6,000	8,000	10,000	
1	Now ill.	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	×	—
2	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	×	—	—	—
3	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	×	—	—	—	—
4	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	×	—	—	—	—	—
5	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	×	—	—	—	—	—
6	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	×	—	—	—	—	—
7	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	×	—	—	—	—	—
8	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	×	—	—	—	—	—	—	—
9	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	—	—	—	—	—	—	No higher dilutions
10	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	—	—	—	—	—	—	Do.
11	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—
12	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—
13	,,	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	No higher dilutions
14	,,	—	—	—	—	—	—	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—
15	,,	—	—	—	—	—	—	—	—	—	—	—	+	—	—	—	—	×	—	—	—	—	—	—
16	,,	—	—	—	—	—	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—
17	,,	—	—	—	—	—	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—
18	,,	—	—	—	—	—	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	—	No higher dilutions
19	,,	—	—	—	—	—	—	—	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—	—
20	,,	—	—	—	—	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—	—	—
21	,,	—	—	—	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—
22	,,	—	—	—	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—	—	—	—
23	,,	—	—	—	—	—	—	—	—	+	—	—	—	—	—	×	—	—	—	—	—	—	—	—
24	,,	—	—	—	—	—	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—

27	,,	—	—	—	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
28	,,	—	—	—	—	—	—	—	+	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—
29	,,	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—
30	,,	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—
31	,,	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	No higher dilutions
32	,,	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Do.
33	,,	—	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—
34	,,	—	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—
35	,,	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	No higher dilutions
36	,,	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Do.
37	,,	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Do.
38	,,	—	—	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—
39	,,	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
40	,,	—	—	—	—	—	+	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
41	,,	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
42	,,	—	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
43	,,	—	—	—	+	—	—	—	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—
44	,,	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
45	,,	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
46	,,	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
47	,,	—	+	—	×	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
48	,,	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	No higher dilutions
49	,,	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Do.
50	,,	—	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Do.
51	,,	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
52	,,	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
53	,,	—	—	—	—	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	No higher dilutions

+ denotes complete sedimentation. × denotes marked, but not complete sedimentation.

TABLE II.—*Recoveries from Malta Fever which have shown Complete Reaction in 10-fold Dilution and over.*

No. of Case.	Period after Fever. Year.	Month.	Dilutions of Serum. 10	20	40	50	60	80	100	150	200	300	400	500	600	800	1,000	1,500	Remarks.
1	—	1	—	—	—	—	—	—	—	—	+	—	—	—	—	×	—	—	—
2	—	1	—	—	—	—	—	—	—	—	+	—	+	—	—	—	—	—	—
3	—	1	—	—	—	—	—	—	—	+	×	—	—	—	—	—	—	—	—
4	—	1	—	—	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—
5	—	1	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—
6	—	1	—	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—	—
7	—	2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—	No higher dilutions.
8	—	2	—	—	—	—	—	—	—	—	+	—	—	—	—	×	—	—	—
9	—	2	—	—	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—
10	—	2	—	—	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—
11	—	2	—	—	—	—	—	—	—	+	×	—	—	—	—	—	—	—	—
12	—	2	—	—	—	—	—	+	—	—	—	—	—	×	—	—	—	—	—
13	—	2	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—
14	—	2	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	No higher dilutions.
15	—	2	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	Do.
16	—	2	—	—	+	—	—	—	—	—	×	—	—	—	—	—	—	—	—
17	—	3	—	—	—	—	—	—	—	—	—	+	—	—	—	—	×	—	—
18	—	3	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	No higher dilutions.
19	—	3	—	—	—	—	—	—	—	—	×	—	—	—	—	—	—	—	—
20	—	3	—	—	—	—	—	—	—	×	—	—	—	—	—	—	—	—	—
21	—	3	—	—	—	—	—	+	—	—	—	—	—	—	×	—	—	—	—
22	—	3	—	—	—	—	—	+	—	—	—	—	×	—	—	—	—	—	—
23	—	3	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	No higher dilutions.
24	—	3	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
25	—	3	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
26	—	3	+	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
27	—	4	—	—	—	—	—	—	—	—	—	—	—	+	—	—	—	×	—
28	—	4	—	—	—	—	—	—	—	—	—	×	—	×	—	—	—	—	—
29	—	4	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—
30	—	4	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—

35	—	6	—	—	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—
36	—	6	—	—	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—
37	—	6	—	—	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—
38	—	6	—	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—
39	—	6	—	—	—	—	—	—	+	—	—	—	—	—	—	—	—	—	No higher dilutions.
40	—	6	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—
41	—	6	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—
42	—	6	—	—	—	—	—	+	×	—	—	—	—	—	—	—	—	—	—
43	—	6	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	No higher dilutions.
44	—	6	—	—	—	+	—	—	—	—	—	—	—	—	—	—	—	—	Do.
45	—	6	—	+	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—
46	—	6	—	—	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—
47	—	6	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
48	—	6	+	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
49	—	6	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	No higher dilutions.
50	—	7	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—
51	—	7	—	—	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—
52	—	8	—	—	—	—	—	+	—	—	—	×	—	—	—	—	—	—	—
53	—	8	—	—	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—
54	—	9	—	—	—	—	—	—	—	—	—	+	—	—	—	—	—	—	No higher dilutions.
55	—	10	+	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
56	1	—	—	—	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—
57	1	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
58	1	2	—	—	—	—	—	+	—	—	×	—	—	—	—	—	—	—	—
59	1	2	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
60	1	4	—	—	—	—	—	—	—	—	×	—	—	—	—	—	—	—	—
61	1	8	—	—	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—
62	1	9	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
63	3	—	—	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
64	4	—	+	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—
65	4	—	+	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
66	5	—	—	—	—	+	—	—	+	—	—	—	—	—	—	—	—	—	—
67	5	—	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
68	7	—	—	+	—	×	—	—	—	—	—	—	—	—	—	—	—	—	—

+ denotes complete sedimentation. × denotes marked, but not complete, sedimentation.

which Durham[1] has figured show no rise before the fifth day after ino lation. This observer further states that the rate of development of agglutinins varies inversely with the amount of the dose. Some exp ments of Wright and Semple bear this out; for instance, in a mon which had been inoculated with half an agar tube of the micrococcus serum showed no reaction until the ninth day after inoculation, whe the serum of a monkey infected with but one loopful of a culture gav reaction on the fifth day.

In the case of man, Aldridge,[2] in the paper to which previous refere has been made, notes his failure to observe the reaction before the f day after the onset of the fever. In two cases which we have inve gated from the onset of the disease and in which this could determined with accuracy, a complete reaction in 400-fold dilution observed in one and in 1,500-fold dilution in the other, both on the fou day of the pyrexia.

In Case 1, mentioned above, the first observation, owing to unavo able causes, was not made till the ninth day after the onset of fe and the twenty-third after accidental inoculation. This observat showed complete sedimentation in 800-fold dilution.

In Case 2, the case of A. E. W., it will be seen from the chart that sedimentation value of his serum had been raised before inoculation to by previous treatment with sterilized cultures. This declined to 20 the third day after inoculation while on the eighth day after the onse the fever there was noted a large increase in the amount of agglutin present in the blood, the sedimentation value being 800. From th data it will be seen that in Malta fever the agglutinating substan appear in the blood comparatively early, much sooner than in average case of typhoid fever. They therefore possess a greater diagno value.

The Persistence of the Agglutinins in the Blood after Recovery.

With a view of estimating the duration of the reaction after recove we have investigated sixty-eight cases at different periods after c valescence, with the following results. In twenty-seven individu examined within six months after convalescence was established average serum dilution in which sedimentation was manifest was fo to be about 350-fold. In eighteen cases examined between six mon and a year after recovery the mean sedimentation value was appro mately 250. In seven cases examined between one and two years a recovery an average value of about 100-fold dilution was determin After two years the reaction has been obtained, in many instan in no higher dilutions than in normal sera; eight out of fourt samples of blood taken from persons who had suffered from the ill from two to eight years previously gave no more marked reaction t

[1] Loc. cit.

[2] Loc. cit.

ıal sera, while the remaining six gave a complete or well-marked nentation in 10-fold dilutions or over. We have only met with one ıple in which the characteristic effects of the serum were apparent longer interval than seven years—this case, examined seven and a years after recovery, gave a complete reaction in 20-fold dilution.

Incubation Period for Man.

'ase 1 and Case 2 mark with accuracy the incubation period. In the er there was an interval of fifteen days between the date of infection the onset of fever, while in the latter this interval was sixteen days. e[1] states that the disease has developed in England in persons left Malta from fourteen to seventeen days previously, while hes[2] has observed instances of the onset of fever eight, ten, and teen days after arrival in Malta. Two cases have come under our e in which the first symptoms began at least from eighteen to ıty days after leaving Malta. From these data it will be seen that ncubation period may vary between eight and twenty days, the usual d being probably about fifteen days.

Geographical Distribution.

Vright and Smith[3] by means of the serum sedimentation test confirm 'act that Bruce and others had long surmised—viz., that Malta fever t confined to the Mediterranean basin. In the publication referred ıese authors tabulated ten cases invalided from India, in which the n reaction gave a sedimentation value of an average of about 300-fold ion. Many of these cases at the time of examination presented the l sequelae of the disease. Since the date of that note ten other ıples have been observed by us in Royal Victoria Hospital, ey. These had been invalided for such diseases as malaria, ic fever, and rheumatism. It is worthy of remark that fourteen of the total of twenty such cases observed came from a ɔaratively small station in the Punjab—namely, Meean-Meer; other places in India where infection has occurred are Calcutta, thu, and Nowshera. One man contracted the disease in Hong- g. This list may be further extended by the following cases, h have all been tested with cultures sent out originally from laboratory. (1) Lieutenant W. Glen Liston, I.M.S., has privately ted to us a case of fever contracted in Secunderabad in the Deccan, erum from which gave a reaction in 80-fold dilution at least with the ɔcoccus Melitensis. (2) Dr. Musser and Dr. Sailer[4] report the case army officer who appears to have contracted the disease in Puerto during the Spanish-American war. Malarial plasmodia were

British Medical Journal, May 18, 1889. [2] Loc. cit.
British Medical Journal, April 10, 1897. (*Vide supra*, pp. 10–11.)
Proceedings of the Pathological Society of Philadelphia, February 1, 1899.

frequently sought for in the blood of this officer, but never found. culture of micrococcus Melitensis was agglutinated by high dilutions the patient's blood. (3) Kretz [1] records the case of a physician who h contracted an obstinate fever in Ajaccio, Corsica. This continued months. After recovery his blood serum agglutinated the micrococ Melitensis in a dilution of 300-fold. 4. The following case, which h recently come under our notice, evidently contracted the disease in t country. Patient was a medical man, aged twenty-six years, who h not been abroad since the age of eight years. From March, 18 till the end of September of the same year he was resident physici in a hospital in Plymouth. On October 1 he assumed the duties junior house physician in the out-patient department at St. Bartholome Hospital, London, where he was employed during the whole of th month. He first began to feel ill in the beginning of Novemb complaining of headache, loss of appetite, lassitude and general malai This condition persisted more or less till December, when it beca complicated by oedema of the feet. The oedema afterwards e tended up the legs. No cardiac lesion could be detected and the uri was normal. Up to this period of the illness he was not confined to be but rested as much as was consistent with the fulfilment of his duti The oedema lasted for about a month, and was succeeded by pain a swelling in certain joints. The hips, knees, ankles, left wrist and le temporo-maxillary joints were all attacked in turn. After the disa pearance of these latter symptoms left orchitis set in and lasted for abo a fortnight. Convalescence was established in March, 1898. On during the period when the joints were affected was the temperature ev taken. At that time it was usually found to be about 102° F. in t evenings. The serum reaction was not tried during the course of t illness, but was tested by us one year and two months after convalescenc It was found then to give complete sedimentation of micrococcus Melite sis in 20-fold dilution and traces up to 50-fold dilution. From the fac stated above we may fairly conclude that this was a case of Malta fev contracted either in Plymouth or in London, although no definite sour of infection can be traced.

Data bearing on the Evolution of the Agglutinins during the Course Malta Fever.

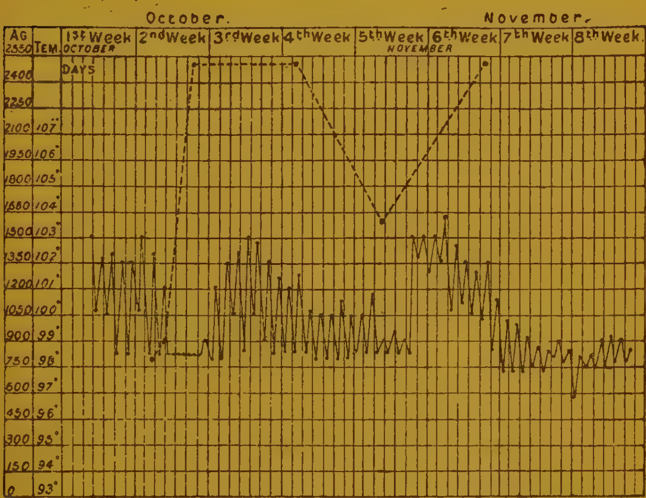
We have seen above that the agglutinating substances which ma their appearance in the blood in cases of Malta fever are specific, and th their action on the micrococcus of Bruce, dead or alive, affords an ea and absolutely trustworthy method of diagnosis between this disea and other fevers which clinically simulate it. We have now to put record the data which we have collected relating to the evolution of the

[1] *Wiener Klinische Wochenschrift*, No. 49, 1897.

tances during the course of the disease. We have already described method which was used in making quantitative estimations of agglu-ıs, and have only to add that in drawing up the agglutination curves with presented the expression which has been adopted for the sedi-ting value of each serum was the dilution in which that serum com-ely sedimented the bacteria with which it was brought in contact, so o leave the supernatant fluid clear. The data were obtained from rvations taken during the course of Malta fever in man. In all en cases were investigated. Seven of these came under observation tically from the beginning of the fever, while the remaining eight brought before our notice during relapses some months after the al attack.

ASE 1.—This is the case of D. S., already referred to. The fever ran mparatively short course and presented no complications. On turn-

D. S. **Chart 1.**



In this and subsequent charts the curve •———•———• represents the temperature, while the curve •- - - - •- - - - • represents the agglutinating power of the serum. This was taken as the highest dilution in which complete sedimentation was observed.

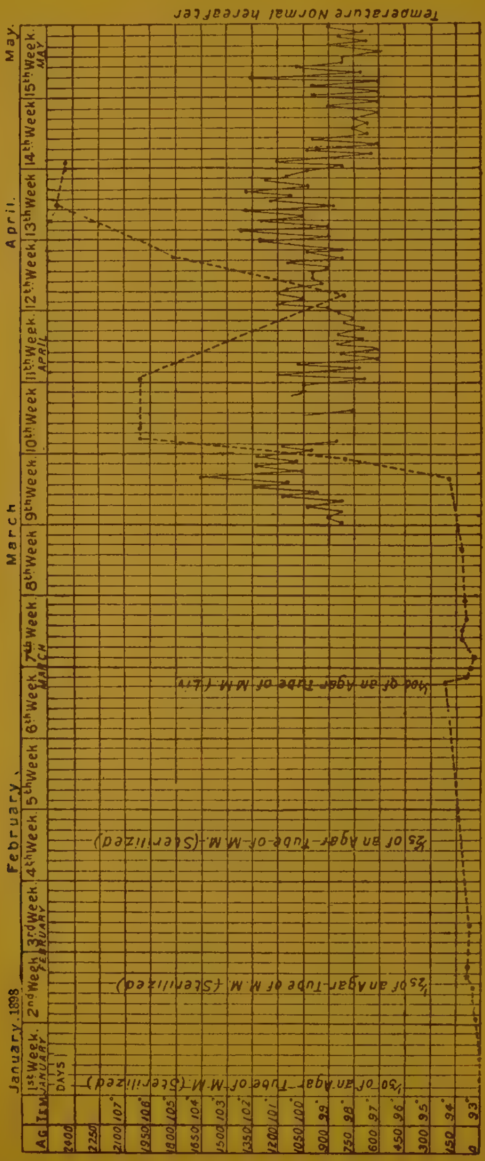
to Chart 1. it will be seen that the agglutinating substances were loped in large quantity soon after the onset of fever, and that, with exception of a slight diminution just before a short relapse, this ıtity was maintained till convalescence was established.

ASE 2.—This is the case of A. E. W., to which reference has already made. The attack was a sharp one, although of comparatively short tion. It was complicated by severe muscular pains, which, however, not retard convalescence. The evolution of the agglutinins in this is of great interest. The salient features of the curve (*vide* Chart 2) are

these: (1) that only a small amount of agglutinins was developed as a result of the three injections of sterilized cultures,[1] which preceded the

CHART 2.

A. E. W.

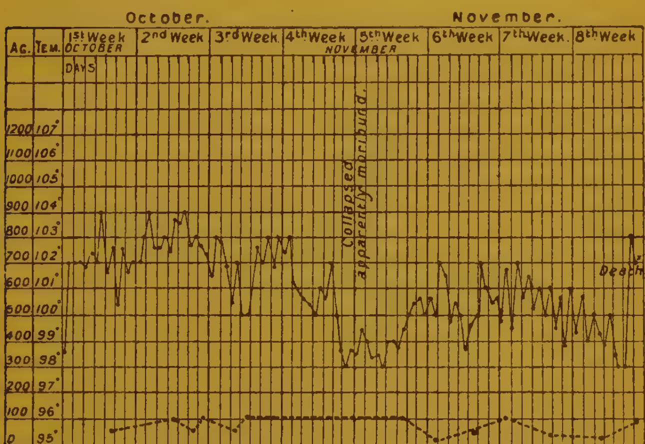


[1] The quantities injected on these occasions were as follows: First injection one-fiftieth of an agar tube (killed at 60° C.); second injection, one twenty-fifth of an agar tube (killed at 60° C.); third injection, one-twenty-fifth of an agar tube (killed at 60° C.); and fourth injection, one-hundredth of an agar tube (living

ulation with the living culture; (2) that a well-marked diminution ne amount of agglutinins followed the inoculation of the living organ-; (3) that a slight and gradual recovery in the amount of agglutinins urred during the period of incubation; (4) that a rapid increase in amount of agglutinins began soon after the onset of the fever; and inally, that, with the exception of a slight fall at the beginning of a idescence of the temperature, this large amount of agglutinins isted till convalescence was well established.

Case 3.—J. T., aged twenty-seven years. Patient had been stationed Ialta for six years, but had never suffered from any fever during this od. Three days before leaving Malta he began to feel lassitude and eral malaise. Pyrexia began on the voyage home. He came under observation about the thirteenth day of the disease. The case was a re one, and presented all the symptoms of an acute specific fever.

J. T. CHART 3.

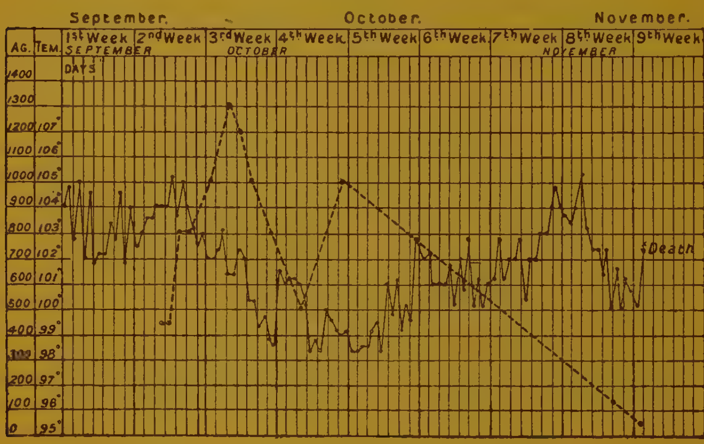


fever was practically continuous till death, which took place about sixty-eighth day of the illness. During the short period of apyrexia n November 3 to 7) the patient was in a collapsed condition and was arently moribund (*vide* Chart 3). The agglutination curve shows throughout the course of the disease the agglutinins were present in blood only in small quantity. The highest dilution which gave com-e sedimentation was 100-fold. The micrococcus Melitensis was ined *post mortem* from the blood and spleen.

Case 4.—G. W., aged twenty-seven years. Patient had served in Malta four months. During this time he had suffered from an attack of nple continued fever"; in other words, the nature of this fever had been diagnosed. He was invalided to Netley at the end of August, 1898. came under our observation in the third week of September, at which

time he had been suffering from fever for about a week. With the exce tion of two very short periods of apyrexia fever was continuous till deat which took place about eight weeks after the beginning of the illne (*vide* Chart 4). The agglutination curve shows : (1) that the agglutini were present in the blood in considerable quantity during the earli period ; (2) that their amount was markedly increased just before ar during the two periods of comparative apyrexia ; and (3) that the substances almost disappeared from the blood shortly before deat Pure cultures of the micrococcus Melitensis were obtained after dea from the blood and spleen.

G. W. CHART 4.



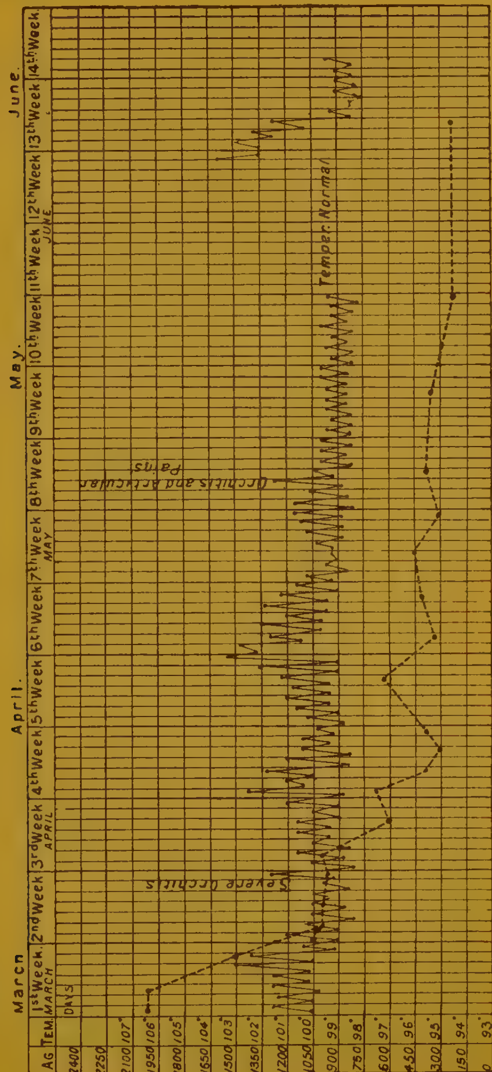
CASE 5.—This is the case of the laboratory attendant, Corporal S., which we have had occasion to refer above. Owing to the patient being c leave he did not come under observation till about the thirty-ninth d of the illness. The case was acute for the first week after admission hospital ; thereupon it became subacute and ultimately it assumed chronic form. Neuralgia and severe double orchitis were present as co plications in the earlier period, and in the later stages severe articul pains accompanied by a return of the orchitis. A relapse supervene upon this (*vide* Chart 5). During the early period of observation the was a very large quantity of agglutinins present in the blood. Th amount diminished markedly at the beginning of the subacute sta and continued to diminish till the chronic relapsing stage was reache Since then they have varied but little. And now, four and a ha months since the onset of the disease, the amount practically remai the same.[1]

[1] *The patient subsequently succumbed to the fever.*

CASE 6.—T. F., aged twenty-six years. Patient had served in Malta eight months. The onset of his illness dates from about eighteen days er leaving Malta and ten days after his arrival in England. The case was te at the outset. After about three weeks the temperature fell to normal.

CHART 5

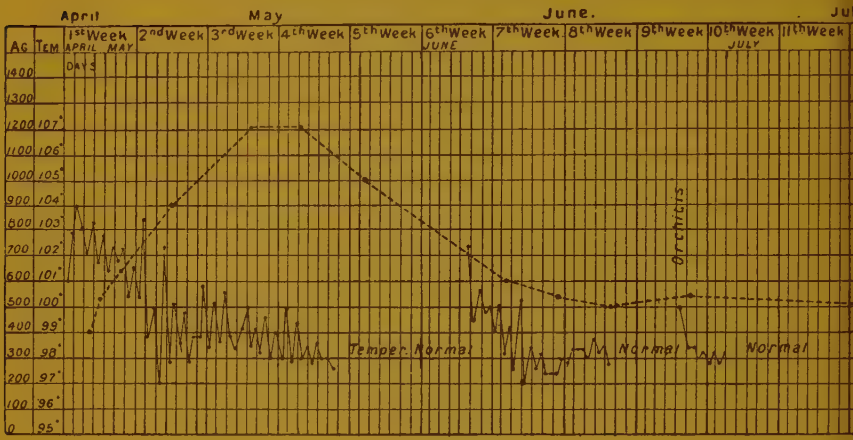
Corporal S.



is defervescence was soon followed by a relapse which lasted for a ek. This was succeeded by an orchitis which was accompanied, however, by little disturbance of temperature. About three weeks after the hitis a second relapse began. This was complicated with swelling of

the submaxillary glands. At the time of writing this relapse still persi
(*vide* Chart 6). The agglutinins were developed gradually in the blo
during the initial stage and had reached a large amount by the ti
the temperature fell. The quantity began to diminish before the ons

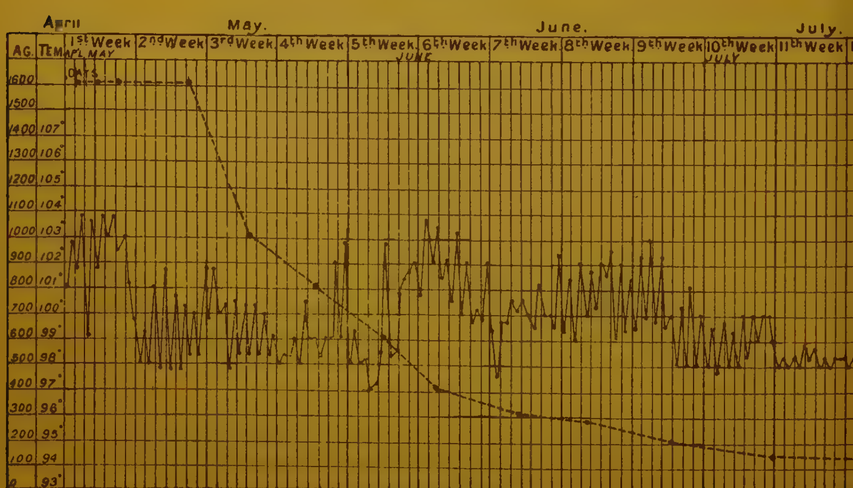
T. F. CHART 6.



of the first relapse, and continued to fall throughout its course. Sin
this the sedimentation curve has varied little up to the present.

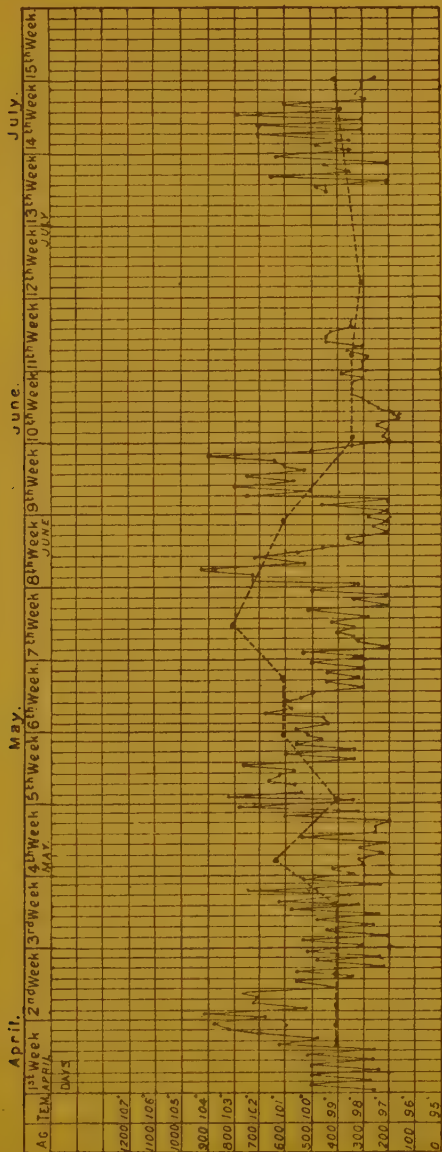
CASE 7.—G. M., aged twenty-two years. Patient had served in Mal
for a year and a half. He had never suffered from any kind of fev

G. M. CHART 7.



g this period. A few days after his arrival at Netley he complained of se, lassitude and loss of appetite; this was soon followed by fever. ame under our direct observation on the fourth day from the com-

M. C. CHART 8.

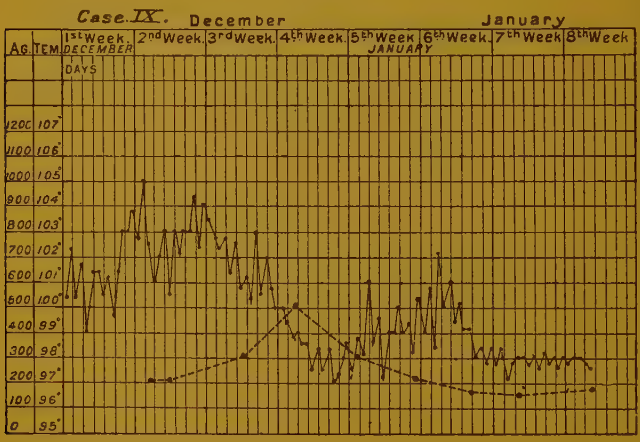


ement of the fever. The case may be compared with that of Case 5, ugh it was more severe. In other words, there was an initial acute , which was followed by a subacute stage interrupted by frequent

relapses. This condition has lasted up to the time of writing, two and half months after its onset (*vide* Chart 7). The agglutinating substan as the curve shows, were present in large quantity during the early peri The amount diminished, at first rapidly, then more slowly in the la stage until it becomes comparatively small. At present it is maintai in this amount.

The remaining eight cases came under our notice at periods vary from two to six months after the onset of the disease. They had b invalided from Malta on account of this malady. They were cases subacute or chronic fever, with periods of apyrexia of variable durati Neuralgia and joint pains were common complications. The quant of agglutinins present in the blood of these patients varied consideral but was never great. In each individual case the agglutinating cu

G. M. CHART 9.



shows distinct rises and falls, more marked in some cases than in oth Two cases are given in detail as types of this class.

CASE 8.—M. C., aged twenty-one years. While in hospita Malta for some slight surgical affection patient suffered from first attack of fever. This lasted for about six weeks, and him very weak and anaemic. Four months after this prim attack and two months after his arrival at Netley the fever recur While the patient was under our observation he suffered from a v marked subacute attack of Malta fever, with occasional periods of hi pyrexia (*vide* Chart 8). This was complicated with neuralgias, j pains and profuse perspirations. The agglutination curve shows t the agglutinins were never present in the blood in very large amoun an average a dilution of about 500-fold causing complete sedimentat

CASE 9.—G. M., aged twenty-seven years. Previously to being inval

patient had suffered in Malta from continued fever for seven weeks. s admitted to Netley about two and a half months from the onset original attack, and about the seventh day of a relapse, which had on the voyage home. This relapse lasted for about three weeks and week of apyrexia was followed by a second relapse, which was, er, less severe and of shorter duration than the first (*vide* Chart 9). gglutination curve shows that the agglutinins during the period review were present in the blood in variable but at no time in large ties. The increase in the amount at the beginning of the apyrexial is worthy of note.

nferences which may be drawn from the Above Observations.

e easiest way of critically sifting the data which have been placed ord above will be to classify the cases in accordance with their l features, thereupon to take up each category of cases separately quire what is the agglutinating curve which corresponds to each ılar category. From the brief sketch of the cases which have been d above it appears that they may be divided into the following l groups :—(1) Severe cases which end fatally within a compara- short time. Case 3 may serve as an example of this category. evere cases which run a more prolonged course, and succumb ne or two relapses. Case 4 and Case 5 may serve as examples of ass. (3) Sharp attacks of fever, which last for a comparatively time, convalescence being rapidly established without any or after t relapse only. These cases have none of the usual complications subacute and chronic cases. Cases 1 and 2 are types of this group. ses which begin acutely, but which become subacute or chronic. ent relapses are usual. They last often for many months, and are cated with orchitis, severe neuralgias, joint pains and profuse ations. They recover eventually. Cases 6, 7, 8, and 9 belong category.

ving thus framed our clinical groups we may now inquire what is ture of the agglutinating curve in each group. In the first group glutinins are present in the blood only in small quantity through- e course of the disease, a 100-fold dilution being the highest in complete sedimentation was observed in the only case in tegory which we have had the opportunity of observing. In ond group the agglutinins may be present in large quantity in the period of the disease, but almost disappear from the blood some efore death. In the case which falls under this category at one time high dilution as 1,300-fold gave complete sedimentation, whereas th the highest dilution giving this reaction was 50-fold. In the group the agglutinating substances are present in large quantity n the disease. The amount remains about constant till convales- s established. The average of the highest dilutions in the cases of

this category was about 2,000-fold. Finally, in the fourth group the agglutinins are found in the blood in large quantity in the initial stage, but fall to a much lower and variable amount during the later stages, the average which then gives complete sedimentation being about 300-fold dilution. The contrast between the evolution of these substances in Group 3 and Group 4 is especially worthy of note.

These results substantiate as far as they go, the inferences which Durham [1] has drawn from a series of observations carried out on rabbits and guinea-pigs infected by means of intra-cerebral inoculation of micrococcus Melitensis. This observer showed that in these animals: (*a*) rapidly fatal infections do not lead to a development of agglutinins in any large quantity in the blood; (*b*) infections in which the animals survive a longer time but ultimately die, may be divided into two groups—(1) those in which the amount of agglutinins, at one time comparatively large, diminishes greatly before death, and (2) those in which the quantity of the agglutinating substances remains comparatively high up to death; and (*c*) infections in which the animals survive may lead to either a great or small production of agglutinins. It will thus be seen that our observations on man coincide with those of Durham on animals in all points but one—namely, that we have no cases to correspond to those constituting Durham's class (*b*) (2), cases, that is, in which the agglutinins were present in comparatively large quantity at death.

Value of an Estimation of the Agglutinating Substances as an Aid to Prognosis.

A cursory study of the cases which have been detailed above and the charts of which are appended will be sufficient to make it apparent that a single estimation of the agglutinins at any one time during the course of an attack of Malta fever would not yield much information as to the severity of the case, nor would it serve as a guide as to its future progress. Further, even a series of observations without an accompanying knowledge of the main clinical features of the case would not justify the observer in forming an opinion as to the probable issue of the case. On the other hand, a series of observations in conjunction with a knowledge of the clinical symptoms would afford much valuable help in arriving at a prognosis. From the data collated above, and from the inferences we have drawn from these data, it would appear to us that the following conclusions are justified: (1) An unfavourable prognosis would be suggested in those cases which are severe from the outset and have a persistently low agglutinating reaction. In such cases the prognosis would no doubt be already indicated by the clinical symptoms, but the fact that the agglutinating power of the blood was low throughout would materially strengthen the clinician in his opinion. (2) We should also be apprehensive of the result in grave attacks in which the agglutination reaction value rapidly falls

[1] Loc. cit.

m a high figure to almost zero. (3) On the other hand, a persistently h and rising agglutination curve sustained well into convalescence uld augur an auspicious and speedy ending to the case notwithstanding acuteness of the symptoms. (4) Finally, a guarded prognosis would m requisite in those cases in which the amount of agglutinins, at first ge, decreases considerably, although the clinical features of the case ght not give rise to any anxiety. In such instances a long illness l one complicated with relapses would be anticipated. It is evident t the estimation of the amount of agglutinins present in the blood is only sure method of deciding whether a case belongs to one or other the last two categories.

It is interesting and important to note that Courmont of Lyons [1] has ived at similar conclusions in regard to typhoid fever. This observer estigated the evolution of the agglutinins in fifty-two cases of typhoid er, and also critically analysed the observations of Widal and Sicard [2] ring on the same problem. His conclusions are as follows: (1) A large antity of agglutinins present in the blood is always favourable (the ger it is the better), especially if this coincides with a remission of the nperature. (2) A small quantity of agglutinins with a rise of temperature always unfavourable. (3) A strong agglutinating power is good at all es in the course of the disease; a low agglutinating power is usually l, as the latter condition is found either in slight forms with a tendency relapse or in very severe cases.

In conclusion, it is our pleasant task to have to thank Professor Wright l Major D. Semple for placing at our disposal the records of many servations and experiments done by them in this laboratory.

[1] Courmont: *Sero Pronostic de la Fièvre Typhoïde*, 1897, and *Presse Médicale*, 2, January 5, 1898.
[2] *Annales de l'Institut Pasteur*, May 25, 1897, p. 403.

Observations bearing on the Question of the Influence which is exerted by the Agglutinins in the Infected Organism.[1]

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The question as to how far the production of specific agglutinating and sedimenting substances stands in relation to the process of immunisation is a question which is manifestly of the highest moment. That there must be some relation between the production of agglutinins and the production of immunity appears certain, not only from the fact that bacteria undergo distortion and become immobilized under the influence of their corresponding agglutinins, but also from the fact that the bacteria in question are inhibited in their growth when they are transferred to a highly agglutinative culture medium. If nothing else had been elicited concerning the agglutinins, these facts would of themselves suffice to show that agglutinins are "*bacteriotropic*"[2] and anti-bacterial substances.

About this particular point, therefore, there would appear to be no controversy. Widely divergent views have, however, been expressed on the question as to how far the agglutinins exert an influence in warding off a bacterial invasion and in restraining the growth of such bacteria as may before the production of agglutinins have established themselves in the interior of the organism. We do not propose here to enter into consideration of all the arguments which have been advanced with the view of showing that no sensible influence in restraining the growth of bacteria is exerted by agglutinins in the organism. We propose here to confine ourselves to the consideration of the argument that the agglutinins must be inoperative *in vivo*, inasmuch as the bacillus typhosus or the micrococcus Melitensis, as the case may be, continues to cultivate itself in the organism long subsequent to the appearance of agglutinins in the blood. We may begin by pointing out that the conclusion that the agglutinins are inoperative *in vivo* is forced upon us only if it can be established for a

[1] Reprinted from the *Lancet*, Dec. 23, 1899.

[2] The term "*bacteriotropic*," which is here employed, is formed upon the analogy of the terms "neurotropic," etc., which have been introduced by Ehrlich. The affix "tropic" as thus used signifies the property of "turning towards" and of entering into chemical composition with.

tainty that the micro-organisms which cultivate themselves in the ;anism are, as a matter of fact, cultivating themselves in the presence of glutinins. Now this is a question which is capable of being resolved a quantitative determination of the amount of agglutinins in the organs which the micro-organisms are cultivating themselves. The observa-ns which we have made in connexion with this question are as follows.

CASE 1.—The patent had a very severe attack of typhoid fever and d during the third week. At the necropsy, held twenty-four hours [1] after ath, the typhoid bacillus was isolated in pure culture from the spleen. grammes of splenic substance were extracted with an equal weight of rmal saline solution. The amount of agglutinating substance in the ar filtrate which was obtained from the above was quantitatively termined in sedimentation tubes by the method which was described one of us in the *British Medical Journal* of February 5, 1897. At the ne time the agglutinating power of the heart blood was determined in e same manner. The results may be tabulated exactly as follows :—

—	Dilutions.					
	4-fold.	6-fold.	12-fold.	16-fold.	20-fold.	100-fold.
um of heart-blood .	Complete	Complete	Complete	Complete	Complete	Incomplete
um of spleen . . .	Trace	Trace	*Nil*	*Nil*	*Nil*	*Nil*

NOTE.—A slight inaccuracy has been admitted into these and all the subsequent ords, inasmuch as the 12 grammes of spleen which were employed for this experi-nt are treated here as if they had been the equivalent of 12 grammes of splenic um. They were in reality the equivalent of only 10 grammes of splenic serum, smuch as the solid substances of the spleen amount to about one-sixth of its ght. The difference is manifestly one which is not worth taking into account view of the considerable differences which are revealed in the tables of results.

CASE 2.—The patient had a typical attack of typhoid fever. He ccumbed about the end of the fourth week. At the necropsy, held enty-four hours after death, the typhoid bacillus was obtained in pure ture from the spleen. The agglutinins in the heart-blood and in the een were estimated in the same manner as in the first case. The lowing were the results :—

—	Dilutions.					
	10-fold.	20-fold.	40-fold.	100-fold.	150-fold.	200-fold.
um of heart-blood .	Complete	Complete	Complete	Complete	Complete	Incomplete
um of spleen . . .	Incomplete	Trace	Trace	*Nil*	*Nil*	*Nil*

[1] In view of the fact that this and all the subsequent observations were made the winter months, when the temperature of the dead house was very low—so

Case 3.—The patient died after a short typical attack of typhoid fever. At the necropsy, which took place twenty hours after death, Peyer's patches were found to be much swollen and congested. They were not ulcerated. The bacillus typhosus was recovered in pure culture from the spleen. The agglutinins were quantitatively estimated (by the same methods as were employed above) in the heart blood, spleen, and in the Peyer's patches. The estimation of the agglutinins in the spleen miscarried through an accident. The results obtained on the heart blood and in the extract from the Peyer's patches are given below :—

—	Dilutions.					
	8-fold.	16-fold.	20-fold.	50-fold.	80-fold.	100-fold.
Serum of heart blood .	Complete	Complete	Complete	Complete	Complete	Incomplete
Serum from Peyer's patches	Incomplete	Traces	*Nil*	*Nil*	*Nil*	*Nil*

Case 4.—The patient suffered from a continued fever. This fever was during life diagnosed as typhoid only by the agglutination reaction, Death occurred suddenly five days after the fall of temperature. At the necropsy, which was conducted twenty-four hours after death, the bacillus typhosus was obtained in pure culture from the spleen, which was absolutely normal in appearance. The agglutinins in the blood and spleen were estimated as above. The following are the results :—

—	Dilutions.				
	4-fold.	8-fold.	16-fold.	20-fold.	50-fold.
Serum of heart blood . .	Complete	Complete	Complete	Complete	Complete
Serum of spleen . . .	Trace	Trace	Trace	*Nil*	*Nil*

—	Dilutions.			
	100-fold.	150-fold.	200-fold.	300-fold.
Serum of heart blood . .	Complete	Incomplete	Incomplete	Traces
Serum of spleen . .	*Nil*	*Nil*	*Nil*	*Nil*

Case 5.—The patient succumbed to a severe attack of Malta fever which ran a comparatively short course. Death took place in the third

low, in fact, that it held in check the growth of all putrefactive micro-organisms—it would appear justifiable to assume that the results which are exhibited above cannot have been due to any *post mortem* cultivation of typhoid bacilli in the spleen. *Vide* also in this connexion note to Observations 5 and 6.

-k of the disease. A necropsy was made twelve hours after death.[1] The -rococcus Melitensis was obtained in pure culture both from heart blood l spleen. It was obtained in small quantities from the former, and relatively large quantities from the latter. The estimation of the -lutinins in heart blood and spleen was carried out in the same manner n previous cases. The results were as under :—

	Dilutions.					
	4-fold.	10-fold.	20-fold.	50-fold.	100-fold.	200-fold
-um of heart blood .	Com-plete	Com-plete	Com-plete	Com-plete	Com-plete	Incom-plete
-um of spleen . . .	Traces	*Nil*	*Nil*	*Nil*	*Nil*	*Nil*

CASE 6.—The patient suffered from a severe and long-continued attack Malta fever. Death took place five and a half months after the com- -ncement of the disease. At the necropsy, conducted eighteen hours after -th, the micrococcus Melitensis was obtained in pure culture from spleen -l heart-blood. The bacteria in the blood were few, while they were -ntiful in the spleen. The estimation of the agglutinins, which was -nducted as above, gave the following results :—

—	Dilutions.						
	5-fold.	10-fold.	20-fold.	50-fold.	100-fold.	200-fold.	400-fold.
-um of heart-blood	Com-plete	Com-plete	Com-plete	Com-plete	Incom-plete	*Nil*	*Nil*
-um of spleen	*Nil*	*Nil*	*Nil*	—	—	—	—

The above observations would appear clearly to establish that the -een in the case of typhoid and Malta fever is much poorer in agglutina- -g substances than the circulating blood. So far as the spleen in typhoid -er is concerned our observations merely confirm what had previously -en established by Courmont in the course of a research which he insti- -ed with a view to determining the distribution of agglutinins in -rious parts of the body. The research in question showed that in nine -ses of typhoid fever examined *post mortem* the agglutinating power the spleen juice was in every case much less than that of the heart -od. This fact, which was arrived at by us independently in following

[1] In view of the fact that the results here and in the next case are similar in points to the results obtained in the case of typhoid fever it is obvious that the -erence which is drawn in Note 2 above is justified. For the results which are -ained here cannot be explained as being the results of a *post-mortem* cultivation the bacteria in the spleen. This interpretation of the results is inadmissible view of the fact that the *post-mortem* examination was here undertaken only -elve hours after death and that we are here dealing with a micro-organism -ich can hardly develop at all at temperatures below 37° C.

up an entirely different line of inquiry, seems to us to acquire a fundamental importance in view of its bearing on the question of the effect exerted by the agglutinins *in vivo*.

We may therefore proceed to consider the general bearing of the conclusion arrived at above, that the micro-organisms of typhoid and Malta fever cultivate themselves in the interior of the infected organism in a medium which is relatively poor in agglutinins. We may consider in this connexion, first, the case in which a relatively large number of bacteria are introduced into the subcutaneous tissue of a man or animal whose blood and lymph contain sensible quantities of agglutinins.

Before we embark upon this question we shall do well to place before ourselves two facts. First, we must note that when we introduce bacteria into an agglutinative medium we effect a corresponding reduction in the agglutinative power of that medium, inasmuch as the agglutinins enter into chemical combination with the bacteria. Secondly, we must note that when the mass effect of the agglutinins in any medium is reduced beyond a certain minimum an agglutinative effect is no longer exerted by that medium. A plasma or lymph whose agglutinative power is reduced below a certain minimum [1] may therefore for our present purposes be regarded as a non-agglutinative fluid.

Keeping these points in view, we are now in position to consider what will probably happen when a large number of bacteria are subcutaneously introduced into an organism which contains agglutinins. Here and throughout this paper we may, in considering the particular case of the agglutinins, bear in mind that we may draw inferences from what happens in the case of these to what will happen in the case of other anti-bacterial substances. It will be manifest that the bacteria when subcutaneously introduced will come in contact in the first instance only with the agglutinins which are contained in that quantum of lymph which occupies the meshes of that portion of the subcutaneous tissue which is the seat of infection. If now the bacteria have been injected in sufficient quantities to abstract from the lymph all, or what for our purpose amounts to all, the agglutinins that lymph will be converted into a non-agglutinative medium. Within "the non-agglutinative envelope" which will thus have been formed such of the bacteria as have not been chemically affected by the agglutinins will be able to cultivate themselves perfectly unchecked, so far at least as any influence on the part of the blood is concerned. It may be urged

[1] The minimum concentration in which a lymph or serum will agglutinate may thus and *à priori* be expected to vary according as the micro-organisms which are exposed to it are virulent or attenuated bacteria. In the case of the former variety of bacteria—i.e., in the case of the variety of bacteria which are most adapted for preserving their vitality in the interior of the animal organism—we must assume that the cohesive forces which hold together the bacterial protoplasm are relatively more powerful. They are therefore more capable of resisting the disruptive force which is exerted on the bacterial protoplasm by the chemical affinity of the agglutinins or certain elements of that protoplasm. This *à priori* deduction is in conformity with the balance of experience, which is, we think, to the effect that a greater concentration of serum is required when virulent bacteria are to be agglutinated.

at, seeing that the agglutinins which have been abstracted will be placed either by the diffusion of others from the blood into the lymph , failing that, by the transudation through the capillary wall of an glutinative lymph, it will very soon come about that the bacteria the tissue will again be exposed to the full *bacteriotropic essure* of the blood. But consideration will show that where the cteria have been introduced in relatively considerable quantities d where the agglutinative power of the blood, is relatively low the pasge of agglutinins outwards from the blood into the lymph will not under dinary circumstances [1] suffice to keep up the agglutinative power of the mph. For agglutinins will be abstracted from the lymph *pari passu* th the increase of the bacteria which will be taking place.

At this stage we may pause for a moment to consider the bearing of ese considerations on the prognosis and therapeutics of local bacterial vasions. First, with regard to prognosis. The prognosis will, in confority with the above considerations, be *pro tanto* good wherever the seat infection becomes hot. For this local rise of temperature and the ight red colouration will be indicative of an increased afflux of arterial ood and of a correspondingly increased lymph-flow. Under conditions ch as these the bacteria will not readily find an opportunity of establishg round themselves a non-bacteriotropic envelope. The prognosis will, the contrary, be serious when local temperature falls, and whenever anosis is developed at the seat of infection. For this will be indicative an arrested or lessened arterial supply and of a consequent stagnation lymph in the tissue. Under these conditions the bacteria will readily able to establish round themselves a non-bacteriotropic envelope and thin this will be able to cultivate themselves unrestrained.

Next with regard to therapeutic measures. The facts which have been nsidered above bring out very clearly that even where a sufficiency of ective anti-bacterial substances are contained in the blood the invading cteria may none the less gain the upper hand if these anti-bacterial bstances are not poured in a continuous stream through the invaded ssue so as everywhere to envelop the invading micro-organisms. This ference is in agreement, not only with the results of experiments on imals, but with the therapeutic principle which has been established

[1] We may remark in passing that if this holds true of the bacteria which lie e in the meshes of the subcutaneous tissue it will *à fortiori* hold true of cteria which have been enclosed in a collodion bag. It does not appear have been sufficiently realized by the observers who have employed these llodion bags in their experiments that a collodion membrane must inevitably, ile it holds back the white blood corpuscles, also restrain the lymph stream ich would otherwise be passing over the bacteria. Now it appears to us quite ssible that it may be this restraint of the lymph stream rather than the holding ck of the white blood corpuscle which is the operative factor in favouring the owth of the bacteria. These same considerations would apply also to the envelopg pellets of agar which were employed by Vaillard in his well-known experiments th tetanus.

by experience in the case of man, that exposure to cold exerts a prejudicial effect by increasing the susceptibility to infection, while the application of heat, either in the form of hot fomentations or in any other form, exerts a beneficial effect on local bacterial invasion. The explanation of this prejudicial influence of cold is to be found in the fact that it arrests the lymph stream and thus permits of the bacteria cultivating themselves in a non-bacteriotropic medium. The contrary good effect of heat is explained by assuming that it increases the lymph stream, and that it does so is shown by its effect in raising a blister.

Having briefly glanced at these questions, we may now turn back and consider what is the bearing of the fact that micro-organisms cultivate themselves in the infected organism in a non-agglutinative medium, upon the phenomena which come under observation when the bacteria, which have introduced them into the subcutaneous tissue, are carried on by the lymph stream into the blood. We may in this connexion with advantage consider what is the probable sequence of events when at the very outset of a typhoid or Malta fever attack the bacteria are carried on into the blood. At the particular stage we may assume that in a person who has not been previously immunised the agglutinins, if they are present at all, will be present only in very small quantities. Consequently we may assume that some at least of the invading bacteria will pass through the blood-stream quite unharmed. These will, in conformity with the general law which obtains when bacteria are introduced in the blood stream, be deposited in the spleen and other internal organs. Having definitely established a lodgment there the bacteria will grow out into colonies, and each of these colonies will as it grows establish round itself a non-agglutinative envelope. As the result of this, and we have seen that the inference has been confirmed, the spleen will as a whole contain less agglutinative substances than the circulating blood. We may further assume that as the fever develops, and as the agglutinins and other anti-bacterial substances are produced in greater quantities, a period will arrive when the agglutinins will be present in the blood in sufficient concentration to permit of their penetrating and abolishing the non-agglutinative envelopes which surround individual colonies. This done, the production of toxins will be arrested and the temperature will fall.

But if when this was effected there happened to remain over somewhere in the organism, shut off, for instance, in a capillary which had become blocked or in some other part which was not freely permeated by the blood stream or the lymph stream, a single bacterial colony, conditions would obviously exist which might afterwards lead on either to a relapse or to secondary local inflammatory process. For the bacteria, sheltered as they would be in the interior of such a *non-bacteriotropic nidus*, might go on cultivating themselves there until they had, as occurs before relapses in typhoid and Malta fever, modified the blood in such a manner as to render it less agglutinative. Such is the mental picture or theory of the typhoid and Malta fever process which is suggested to us by our

servations. We have endeavoured to test the theory by controlling, far as opportunity offered, the deductions which can be drawn from this eory by actual observations. With a view to doing this we endeavoured determine whether the characteristic distribution of the bacillus phosus in the infected organism (absence of the bacillus from the circu-ing blood and growth of the bacillus in discrete colonies in the spleen d other internal organs and in the typhoid spots) can be satisfactorily plained in conformity with the theory. The first point which attracts ention here is the absence of the bacillus typhosus in blood and the owth of that bacillus in the spleen in the form of discrete colonies. As have seen from the comparative estimation of the agglutinins in the ood and spleen, this distribution accords well with the assumption that e agglutinins are an effective factor in checking the growth of the bacillus. however, appeared desirable further to test the correctness of this hypo-esis, and it appeared that light might be thrown upon the question by termining whether in those septicaemic diseases in which bacteria ordi-rily run riot all over the organism the bacteria would, after the intro-ction of anti-bacterial substances, be found to survive only in the form discrete colonies in the organs. Opportunities for determining this int presented themselves in connexion with certain experiments which re made by one of us in conjunction with Lieutenant W. Glen Liston, I.S., on the effects of the introduction of Roux's anti-plague serum o the organism of plague-infected guinea-pigs. It was determined in e experiments in question that in the case of animals which had received ficient quanta of serum such plague bacilli as survived were to be found ly in discrete colonies in the organs. In the untreated control guinea-gs the bacilli were, as usual, found in countless numbers in the circula-g blood and all over the body. It would appear that the hypothesis unciated above wins support from these observations. With regard the question as to whether the cultivation of the bacillus typhosus in e typhoid spots was or was not, as in the case of the cultivation of this cillus in the spleen, to be regarded as a cultivation which is taking place a non-agglutinative medium, it seemed possible to learn something making comparative estimations of the agglutinating power of the ood from the finger-tip and of the mixture of blood and lymph which n be obtained by puncturing and pressing the typhoid spots. We have d only three opportunities of applying this test to the theory. In each se we made a comparative estimation of the agglutinating power of the serum derived from a typhoid spot, and (*b*) the serum derived in e ordinary way from the finger-tip of the patient in question. The ult of these comparative estimations (made in capillary sedimentation bes by methods employed above) were as follows :—

CASE 1.

	Dilutions.				
	12-fold.	24-fold.	48-fold.	96-fold.	192-fold.
Serum from typhoid spots	Incomplete but marked	Incomplete but marked	Trace	*Nil*	*Nil*
Serum from finger-tip .	Complete	Nearly complete	Incomplete but very well marked	Incomplete but very well marked	Incomplete but distinct

CASE 2.—*Child, beginning of the Second Week of Fever; Delirious; Copious Eruptions of Rose Spots: very marked Improvement supervened twenty-four hours after.*

	Dilutions.						
	12-fold.	24-fold.	48-fold.	96-fold.	192-fold.	384-fold.	768-fold.
Serum from typhoid spots	Reaction marked	Marked	Trace	*Nil*	*Nil*	*Nil*	*Nil*
Serum from finger-tip . .	Reaction complete	Complete	Complete	Complete	Complete	Complete	Trace

CASE 3.—*Child; beginning of the Second Week of Fever; Mild Attack; Copious Eruptions.*

	Dilutions.				
	12-fold.	24-fold.	48-fold.	96-fold.	192-fold.
Serum from typhoid spots	Mere trace	Mere trace	Mere trace	*Nil*	*Nil*
Serum from finger-tip	Complete	Complete	Complete	Trace	*Nil*

It will be seen that these observations, so far as they go, confirm the correctness of the theory enunciated.

n the Bactericidal Effect exerted by Human Blood on certain Species of Pathogenic Micro-organisms,

And on the Anti-bactericidal Effects obtained by the Addition to the Blood in vitro of Dead Cultures of Micro-organisms in Question [1]

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Data with regard to the Bactericidal Power of the Blood as affecting the Bacillus Typhosus and Spirillum Cholerae Asiaticae—II. Data with regard to the Bactericidal Power of the Blood as affecting the Staphylococcus Pyogenes—III. Data with regard to the Bactericidal Power of the Blood as affecting the Bacillus Pestis—IV. Data with regard to the Bactericidal Power of the Blood and the Micrococcus Melitensis—Conclusions.

E fact that the blood of ordinary laboratory animals exerts a very rked bactericidal effect upon the *Bacillus typhosus* and the *Spirillum lerae asiaticae*, while it exerts little or no effect upon the *Staphylococcus* 1 *Streptococcus pyogenes*, has hardly received the attention which would seem to merit, in view of the circumstance that these facts olve the important problem as to whether the blood exerts its bacter- dal action upon pathogenic organisms generally, or only upon certain cies of such micro-organisms.

We have addressed ourselves to the task of re-investigating this eral problem by the aid of the methods of bactericidal estimation ich have been elsewhere described by one of us,[2] conducting our eriments upon human blood, and drawing within the sphere of our servation, not only the micro-organisms particularized above, but o the micrococcus Melitensis of Bruce and the *bacillus pestis*.

Data with regard to the Bactericidal Power of the Blood as affecting the Bacillus Typhosus and Spirillum Cholerae Asiaticae.

We may begin by setting forth certain data in connexion with the cteridical power of human blood upon the *bacillus typhosus*, and

[1] Reprinted from the *Journal of Hygiene*, Vol. II., No. 4, Oct., 1902.

[2] *Lancet*, June 1, 1901, p. 1,532; *Proc. Roy. Soc.*, Vol. 71.

TABLE I.—*Exhibiting* (a) *the Bacterial Action exerted by Human Serum on Bacillus Typhosus, and* (b) *the Anti-bactericidal Effect obtained by the Addition of a Sterilized Typhoid Culture to the Mixture of Serum and Living Culture.*

Dilutions in which the Living Typhoid Culture was employed.	Capillary Testing Pipettes were filled with Mixture of											
	2 vols. F. N. W.'s Serum, 1 vol. Dilution of Living Culture which contained 33,000,000 *T.B.* per c.c., and		1 vol. H. B.'s Serum, 1 vol. Dilution of Living Culture which contained 220,000,000 *T.B.* per c.c., and		2 vols. A. E. W.'s Serum, 1 vol. Dilution of Living Culture which contained 210,000,000 *T.B.* per c.c., and		1 vol. H. B.'s Serum, 1 vol. Dilution of Living Culture which contained 220,000,000 *T.B.* per c.c., and		1 vol. A. E. W.'s Serum, 1 vol. Dilution of Living Culture which contained 260,000,000 *T.B.* per c.c., and		1 vol. A. E. W.'s Serum, 1 vol. Dilution of Living Culture which contained 540,000,000 *T.B.* per c.c., and	
	1 vol. Sterile Broth.	1 vol. Sterilized Typhoid Culture.	1 vol. Sterile Broth	1 vol. Sterilized Typhoid Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Culture which contained 210,000,000 *T.B.*	1 vol. Sterile Broth	1 vol. Sterilized Dead Typhoid Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Culture which conta ned 260,000,000 *T.B.*	1 vol. Sterile Broth.	1 vol. Sterilized Typhoid Culture.
Undiluted . .	Growth	—	Growth	—	Growth	—	Growth	Growth	Growth	Growth	Growth	Growth
2-fold dilut.	,,	Growth	,,	Growth	,,	Growth	,,	,,	,,	,,	,,	,,
5 ,, ,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
10 ,, ,,	Sterile	,,	,,	,,	Sterile	,,	,,	,,	,,	,,	Sterile	,,
25 ,, ,,	,,	,,	—	—	,,	,,	,,	,,	,,	,,	Growth	,,
50 ,, ,,	,,	,,	Sterile	,,	,,	,,	Sterile	,,	Sterile	,,	Sterile	,,
100 ,, ,,	,,	,,	,,	,,	,,	Sterile	,,	,,	,,	,,	,,	,,
1,000 ,, ,,	,,	Sterile	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
10,000 ,, ,,	,,	,,	,,	Sterile	,,	,,	,,	Sterile	,,	,,	,,	,,
100,000 ,, ,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,

F. N. W. and H. B. were normal men. A. E. W. had nine months previously, and also on previous occasions, undergone anti-typhoid inoculation.

In every case, both in this and in the subsequent tables, the serum was employed within two or three hours after the blood had been drawn off.

In every case, both in this and in the subsequent tables, the living cultures employed were, unless otherwise stated, twenty-four hour old broth cultures.

Here and in the subsequent tables the sterilized bacterial cultures employed had been sterilized by exposure to a temperature of about 60°–65° C. for ten to twenty minutes.

Here and in the subsequent tables the sera were allowed to act upon the bacteria with which they were mixed for eighteen to twenty-four hours at 37° C. before the effect was tested.

In every case the term "growth" denotes that the growth obtained presented the characters of a pure cultivation of the micro-organism employed in the test. In cases of doubt the purity of the culture was tested by subcultures and microscopical examination. Where contaminations were found the series of experiments was in almost every instance rejected. Where such a series is retained

TABLE II.—*Exhibiting the Anti-bactericidal Effects produced by the Addition to a Mixture of Serum and Living Typhoid Culture of Filtrates from Typhoid Cultures.*

Dilutions in which the Living Typhoid Culture was employed.	Capillary Testing Pipettes were filled in with: 1 vol. F. N. W.'s Serum, 1 vol. Dilution of Living Culture which contained 76,000,000 *T.B.* per c.c. and				1 vol. A. E. W.'s Serum, 1 vol. Dilution of living Culture which contained 76,000,000 *T.B.* per c.c., and		
	1 vol. Sterile Broth.	1 vol. Filtrate from a 24 hours Typhoid Culture.	1 vol. Filtrate from a 5 months old Typhoid Culture.	1 vol. same 5 months old Typhoid Culture unfiltered.	1 vol. Sterile Broth.	1 vol. Filtrate rom a 24 hour Typhoid Culture.	1 vol. Filtrate from a 5 months old Typhoid Culture.
2-fold dilution .	Growth	Growth	Growth	Growth	Sterile	Growth	Growth
5 ,, ,,	Sterile [1]	,,	,,	,,	,,	,,	,,
10 ,, ,,	Growth	,,	,,	,,	,,	Sterile	,,
25 ,, ,,	Sterile	Sterile	,,	,,	,,	,,	Sterile
50 ,, ,,	,,	,,	,,	,,	,,	,,	,,
100 ,, ,,	,,	,,	,,	,,	,,	,,	,,
1,000 ,, ,,	,,	,,	Sterile	Sterile	,,	,,	Contamination [2]
10,000 ,, ,,	,,	,,	,,	,,	,,	,,	Sterile
100,000 ,, ,,	,,	,,	,,	,,	,,	,,	,,
1,000,000 ,, ,,	,,	,,	,,	,,	,,	,,	—

With regard to F. N. W. and A. E. W. and general condition of the experiments, see notes to Table I.

[1] Sterility of the tube probably due to accidental overheating.

[2] Staphylococcus.

with regard to the "anti-bactericidal effect" obtained by the introduction of a sterilized culture of the typhoid bacillus into human blood *in vitro*.[1]

A point of incidental interest here suggests itself in connexion with the question as to what is the element in the sterilized culture which exerts the anti-bactericidal effect exemplified in Table I.

The experiments subjoined in Table II are typical examples of a number of experiments instituted with a view to the determination of this question.

These results show that a filtrate from a young culture of B. typhosus exerts little or no anti-bactericidal effect; while a filtrate from an old culture which contains in solution elements derived from the dissolution of the typhoid bacilli exerts a very marked anti-bactericidal effect. Of particular interest are the results in columns 3 and 4, which show that the filtrate derived from a culture in which the bacilli had been macerating at 37° C. for a period of five months, diminished the bactericidal power of the serum with which it was mixed to exactly the same degree as the unfiltered culture.

Passing to the consideration of the bactericidal effect exerted by human serum upon the cholera vibrio, we subjoin a selection of typical experiments illustrating on the one hand the bactericidal effect exerted upon the cholera vibrio, and on the other hand, the diminution of bactericidal power which is achieved by the addition of a sterilized cholera culture to a mixture of serum and living cholera culture.

It will be manifest from a comparison of the experiments in Table I and Table III that the bactericidal and anti-bactericidal effects proceed on precisely the same lines whether we are employing a culture of typhoid or a culture of cholera.

It becomes, therefore, a point of interest to determine whether a diminution of the bactericidal effect exerted on the typhoid bacillus is obtained by the addition of a sterilized cholera culture to the mixture of serum and living typhoid culture; and *vice versâ* whether a diminution of the bactericidal effect exerted on the cholera vibrio is obtained by the addition of a sterilized typhoid culture to a mixture of serum and living cholera vibrios.

Tables IV and V, which show the effect invariably obtained in our experiments, supply the answer to this question.

As shown in the Tables IV and V, taken in conjunction with Tables I and III, the anti-bactericidal effect which is in each case obtained, is obtained indifferently with either variety of sterilized culture. We

[1] Data with regard to the first of these points have already been set forth by one of us in a paper published in the *Lancet*, September 14, 1901, p. 715, dealing with the changes produced in the blood by anti-typhoid inoculation. The second of these questions has also been briefly adverted to in the same journal, June 1, 1901, p. 1534, in connexion with a suggestion that the anti-bactericidal effect exerted might serve as a criterion for the standardization of bacterial vaccines.

TABLE III.—*Exhibiting* (a) *the Bactericidal Effect exerted by Serum on a 24-hour old Culture of the Cholera Vibrio, and* (b) *the Anti-bactericidal Effect achieved by the addition of a Sterilized Cholera Culture to the Mixture of Serum and Living Cholera Culture.*

Dilutions in which the Living Cholera Culture was employed.	Capillary Tubes were filled with							
	1 vol. F. N. W.'s Serum 1 vol. Dilution of Living Culture which contained 240,000,000 Cholera Vibrios per c.c., and		1 vol. A. E. W.'s Serum, 1 vol. Dilution of Living Culture which contained 45,000,000 Cholera Vibrios per c.c., and		1 vol. Serum of Rabbit No. 1, 1 vol. Dilution of Living Culture which contained 45,000,000 Cholera Vibrios per c.c., and		1 vol. Serum of Rabbit No. 2, 1 vol. Dilution of Living Culture which contained 45,000,000 Cholera Vibrios per c c., and	
	1 vol. Sterile Broth.	1 vol. Sterilized Cholera Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Cholera Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Cholera Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Cholera Culture.
Undiluted culture . . .	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth
2-fold dilution . .	,,	,,	Sterile	,,	,,	,,	Sterile	,,
5 ,, ,, . .	,,	,,	,,	,,	,,	,,	,,	,,
10 ,, ,, . .	Sterile	,,	,,	,,	,,	,,	,,	,,
25 ,, ,, . .	,,	,,	,,	,,	Sterile	,,	,,	,,
50 ,, ,, . .	,,	,,	,,	,,	,,	,,	,,	,,
100 ,, ,, . .	,,	,,	,,	,,	,,	,,	,,	Sterile
1,000 ,, ,, . .	,,	,,	,,	,,	,,	,,	,,	,,
10,000 ,, ,, . .	,,	,,	,,	Sterile	,,	,,	,,	,,
100,000 ,, ,, . .	,,	,,	,,	,,	,,	Sterile	,,	,,

With regard to F. N. W. and A. E. W. see notes to Table I.

Rabbit 1 had been inoculated with one tube of typhoid bacillus.

Rabbit 2 ,, ,, ,, ,, ,, cholera vibrio.

Table IV.—*Exhibiting* (a) *the Bactericidal Effect exerted on a Typhoid Culture, and* (b) *the Diminution of that Effect which is achieved by the Addition of a Sterilized Cholera Culture.*

Dilutions in which the Living Typhoid Culture was employed.	Capillary Tubes were filled with							
	1 vol. F. N. W.'s Serum, 1 vol. Dilution of 24-hour old Living Broth Culture of the Typhoid Bacillus, and		1 vol. A. E. W.'s Serum, 1 vol. Dilution of 24-hour old Living Broth Culture of the Typhoid Bacillus, and		1 vol. Serum of Rabbit 1, 1 vol. Dilution of a 24-hour old Living Broth Culture of the Typhoid Bacillus, and		1 vol. Serum of Rabbit 2, 1 vol. Dilution of 24-hour old Living Broth Culture of the Typhoid Bacillus, and	
	1 vol. Sterile Broth.	1 vol. Sterilized Cholera Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Cholera Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Cholera Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Cholera Culture.
Undiluted culture . . .	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth
2-fold dilution . .	”	”	”	”	”	”	”	”
5 ” ” . .	”	”	”	”	”	”	”	”
10 ” ” . .	Sterile	”	”	”	Sterile	”	”	”
25 ” ” . .	”	”	Sterile	”	”	”	”	”
50 ” ” . .	”	”	”	”	”	”	”	”
100 ” ” . .	”	”	”	”	”	”	”	”
1,000 ” ” . .	”	”	”	”	”	”	”	”
10,000 ” ” . .	”	”	”	”	”	”	Sterile	”
100,000 ” ” . .	”	Sterile	”	Sterile	”	”	”	”
1,000,000 ” ” . .	—	—	”	”	”	”	”	”

With regard to F. N. W., A. E. W., and Rabbits 1 and 2, see notes to Table III.

TABLE V.—*Exhibiting* (a) *the Bactericidal Effect exerted on a Cholera Culture, and* (b) *the Diminution of that Effect produced by the Addition of a Sterilized Typhoid Culture.*

Dilutions of the Living Cholera Culture which were employed.	Capillary Testing Pipettes were filled with: 1 vol. A. E. W.'s Serum, 1 vol. Dilution of Living Cholera Culture which contained 466,000,000 Cholera Vibrios per c.c., and		1 vol. Serum of Rabbit 1, 1 vol. Dilution of Living Cholera Culture which contained 85,000,000 Cholera Vibrios per c.c., and		1 vol. Serum of A. E. W., 1 vol. Dilution of Living Cholera Culture, and		1 vol. W. G. L.'s Serum, 1 vol. Dilution of Living Cholera Culture containing 2,600,000 Cholera Vibrios per c.c. and	
	1 vol. Sterile Broth.	1 vol. Sterilized Typhoid Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Typhoid Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Typhoid Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Typhoid Culture.
Undiluted culture . . .	Growth	Growth	Growth	Growth	Sterile	Growth	Growth	Growth
2-fold dilution . .	"	"	"	"	"	"	"	"
5 " " . .	"	"	"	"	"	"	"	"
10 " " . .	"	"	"	"	"	"	"	"
25 " " . .	Sterile	"	Sterile	"	"	"	"	"
50 " " . .	"	"	"	"	"	"	"	"
100 " " . .	"	"	"	"	—	"	"	"
1,000 " " . .	"	"	"	"	—	"	Sterile	"
10,000 " " . .	"	"	"	"	—	Sterile	"	Sterile
100,000 " " . .	"	"	"	"	—	—	—	"

With regard to A. E. W. and Rabbit No. 1, see notes to Table III.

TABLE VI.—*Exhibiting the Results of the Blood Examinations carried out on Two Rabbits which were Inoculated with similar quantities, in the one case of a Sterilized Typhoid Culture, in the other case of a Sterilized Cholera Culture. These Rabbits are in the table below denoted respectively as the Typhoid Rabbit and the Cholera Rabbit.*

Dilutions of the Living Cultures which were employed in the testings.	Tests carried out immediately antecedent to Inoculation.				Tests carried out 24 hours after Inoculation.			
	Capillary Testing Pipettes were filled in with				Capillary Testing Pipettes were filled in with			
	1 vol. Dilution of a Culture of Typhoid which contained 94,000,000 *T.B.* per c.c., and		1 vol. Dilution of a very thin Culture of Cholera and		1 vol. Dilution of a Culture of Typhoid which contained 446,000,000 *T.B.* per c.c., and		1 vol. Dilution of a very thin Culture of Cholera and	
	1 vol. Serum of Typhoid Rabbit.	1 vol. Serum of Cholera Rabbit.	1 vol. Serum of Typhoid Rabbit.	1 vol. Serum of Cholera Rabbit.	1 vol. Serum of Typhoid Rabbit.	1 vol. Serum of Cholera Rabbit.	1 vol. Serum of Typhoid Rabbit.	1 vol. Serum of Cholera Rabbit.
Undiluted culture . . .	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth
2-fold dilution . .	"	"	Sterile	Sterile	"	"	Sterile	"
5 " " . .	"	"	"	"	"	"	"	Sterile
10 " " . .	"	"	"	"	Sterile	"	"	"
25 " " . .	"	Sterile	"	"	"	"	"	"
50 " " . .	Sterile	"	"	"	"	"	"	"
100 " " . .	"	"	"	"	"	"	"	"
1,000 " " . .	"	"	"	"	"	"	"	"
10,000 " " . .	"	"	"	"	"	Sterile	"	"
100,000 " " . .	"	"	"	"	"	"	"	"

TABLE VI—(*continued*).

Dilutions of the Living Cultures which were employed in the testings.	Tests carried out 48 hours after Inoculation.				Tests carried out 14 days after Inoculation.			
	Capillary Testing Pipettes were filled in with				Capillary Testing Pipettes were filled in with			
	1 vol. Dilution of a Culture of Typhoid which contained 324,000,000 *T.B.* per c.c., and		1 vol. Dilution of a Culture of Cholera which contained 400,000 *C.V.* per c.c., and		1 vol. Dilution of a Culture of Typhoid which contained 480,000,000 *T.B.* per c.c., and		1 vol. Dilution of a Culture of Cholera which contained 85,000,000 *C.V.* per c.c., and	
	1 vol. Serum of Typhoid Rabbit.	1 vol. Serum of Cholera Rabbit.	1 vol. Serum of Typhoid Rabbit.	1 vol. Serum of Cholera Rabbit.	1 vol. Serum of Typhoid Rabbit.	1 vol. Serum of Cholera Rabbit.	1 vol. Serum of Typhoid Rabbit.	1 vol. Serum of Cholera Rabbit.
Undiluted culture . . .	Growth	Growth	Growth	Sterile	Growth	Growth	Growth	Sterile
2-fold dilution . .	„	„	„	„	„	„	„	„
5 „ „ . .	Sterile	„	Sterile	„	„	„	„	„
10 „ „ . .	„	„	„	„	Sterile	„	Sterile	„
25 „ „ . .	„	„	„	„	„	„	„	„
50 „ „ . .	„	„	„	„	„	„	„	„
100 „ „ . .	„	„	„	„	„	„	„	„
1,000 „ „ . .	„	„	„	„	„	Sterile	„	„
10,000 „ „ . .	„	Sterile	„	„	„	„	„	„
100,000 „ „ . .	„	„	„	„	„	„	„	„

must consequently assume either that the bactericidal substance in the serum which kills the typhoid bacillus is one and the same substance which kills the cholera vibrio, or alternatively, that the bactericidal substance which kills the cholera vibrio possesses an element in common with the bactericidal substance which kills the typhoid bacillus.

With a view to deciding between these alternatives, we have investigated the question as to whether the inoculation of a full dose of anti-typhoid vaccine, which produces in man a preliminary diminution and subsequent increase in the bactericidal effect exerted on the typhoid bacillus,[1] brings about any similar diminution and increase in the bactericidal effect exerted upon the cholera vibrio.

The following observations bear on the question.

The bloods of three healthy men, who recently came up for prophylactic inoculation with anti-typhoid vaccine, were tested before inoculation, and afterwards, at intervals of a few days, against both the typhoid bacillus and the cholera vibrio. In no case was any indication obtained of an alteration in the bactericidal effect exerted on the cholera vibrio, although the negative and positive phases of diminished and exalted bactericidal power with respect to the typhoid bacillus manifested themselves in a typical manner.

These results confirm those obtained by one of us on two previous patients.

We further investigated the point upon two rabbits inoculated respectively with sterilized cultures of cholera and typhoid.

The results of the blood examinations here made are subjoined in tabular form (Table VI).

A comparison of the first and second testings of the cholera-inoculated rabbit[2] would seem to suggest that an initial reduction of the bactericidal power was exerted upon both species of micro-organisms. It would, in other words, seem to point to the comparability of the immediate effect exerted by the introduction of a sterilized culture of cholera into the animal organism with the effect exerted by the direct introduction of the culture into the serum *in vitro*.

On the other hand, a comparison of the results obtained in the first and last testings of both the typhoid and the cholera-inoculated rabbit will show that the increase of the bactericidal power which was achieved by inoculation was, in each case, an increase only with respect to the particular species of micro-organism which had been inoculated.

The latter datum is for our present purpose the essentially important one of the experiment. It seems to indicate clearly that the bacteri-

[1] Wright, *Lancet*, September 1, 1900, p. 715.

[2] The circumstance that a positive phase of increased bactericidal power was obtained in case of the typhoid rabbit without the intervention of a negative phase of diminished bactericidal power is in accordance with what occurs in man after the inoculation of a relatively small dose of typhoid vaccine (Wright, *Lancet*, September 14, 1901, p. 715).

cidal effects of a serum, at any rate in the case of a serum derived from the immunised animal, is, as is assumed by the theories of Ehrlich and Bordet respectively, achieved by the co-operation of two bactericidal elements, one of these being a chemical agent which exerts an action on more than one species of micro-organism, and the other a chemical agent which is specific for each particular species of micro-organism.

There is, however, nothing to forbid our explaining the bactericidal action of normal serum by the more simple assumption that the non-specific element referred to above ("complement" of Ehrlich, "alexin" of Bordet) suffices by itself to exert a bactericidal effect.

From the study of the action of the serum upon the typhoid bacillus and the cholera vibrio, we pass to the consideration of the action of the serum upon the staphylococcus pyogenes.

II. Data with regard to the Bactericidal Power of the Blood as affecting the Staphylococcus pyogenes.

As a preliminary to setting forth our results, we may observe that we have not in our numerous experiments found any difference of behaviour as between the different varieties of the staphylococcus pyogenes. For this reason we have thought it unnecessary to encumber the tables given below by specifying in each case the particular variety of staphylococcus employed. Suffice it to say that these were chiefly cultures of the staphylococcus aureus and albus freshly cultivated from operation wounds, furuncles and sycosis.

We set forth first a series of typical experiments conducted by mixing in capillary testing pipettes in each case one volume of serum and one volume of a progressively increasing dilution of a twenty-four-hour old staphylococcus culture.

It will be manifest that the results set forth in Table VII are in conformity with the results obtained with the blood of animals in the classical researches of Nuttall.[1] They show that human serum does not exert any bactericidal effect whatever upon the staphylococcus; nay, more, they suggest, and this suggestion is confirmed by direct observation on the colonies grown[2] in capillary testing pipettes filled with equal volumes of serum and gelatine cultures of staphylococcus, that additions of serum exert a favourable influence on the growth of this germ.

Not obtaining any indication of a bactericidal effect exerted in the case of the volume for volume mixture of serum and broth dilutions of staphylococcus cultures, we experimented further, using dilutions of broth cultures made with the serum under examination. In the higher dilutions thus obtained, we are in point of fact dealing with practically undiluted serum.

[1] *Zeitschrift f. Hygiene*, 1888, vol. iv. pp. 353–394.

[2] The technique employed in connexion with the observations here in question was that which was described by one of us in the *Lancet*, December 1, 1900, pp. 1556–1560.

Table VII.—*Exhibiting the Results obtained on Cultivating a Mixture of equal volumes of Serum and of a Graduated Dilution of Staphylococcus Culture which had remained in contact for 18–24 hours at 37° C.*

Dilutions in which the Living Staphylococcus Culture was employed.	Capillary Testing Pipettes were filled with 1 vol. Dilution of Staphylococcus Culture and				
	1 vol. Sterile Broth.	1 vol. F. N. W.'s Serum.	1 vol. A. E. W.'s Serum.	1 vol. W. B. L.'s Serum.	1 vol. J. A.'s Serum.
10-fold dilution	Growth	Growth	Growth	Growth	Growth
100 " "	"	"	"	"	"
1,000 " "	"	"	"	"	"
10,000 " "	"	"	"	"	"
100,000 " "	"	"	"	"	"
1,000,000 " "	"	"	"	"	"
10,000,000 " "	Sterile	"	"	"	"

With regard to F. N. W., A. E. W., and the general conditions of the experiments, *see* notes to Table I. W. B. L. was a man in normal health. J. A. had suffered from furunculosis and sycosis barbae for a period of nine years, and had completely recovered after three successive inoculations of a sterilized culture of staphylococcus aureus cultivated from his boils.

Table VIII.—*Exhibiting the Results of the Cultivations undertaken in the case of Staphylococcus Cultures diluted* (a) *with Sterile Broth, and* (b) *with Undiluted Serum.*

Dilutions of the Cultures which were employed.	6 day Broth Culture of Staphylococcus		2 day Broth Culture of Staphylococcus		
	diluted with Sterile Broth, then transferred to Nutrient Agar and Incubated.	diluted with A. E. W.'s Serum, digested with this 24 hours at 37° C., and then cultivated in Nutrient Broth.	diluted 1,000,000-fold with Sterile Broth, then transferred to Nutrient Agar and cultivated.	diluted with A. E. W.'s Serum, digested with this 24 hours at 37° C., and then cultivated in Nutrient Broth.	diluted with M. G.'s Serum, digested with this 24 hours at 37° C., and then cultivated in Nutrient Broth.
10-fold dilut.	—	Growth obtained from circ. 10 cmm.	—	Growth obtained from circ. 5 cmm.	Growth obtained from circ. 5 cmm.
100 ,, ,,	—	Growth obtained from circ. 10 cmm.	9 colonies from 25 cmm.	Growth obtained from circ. 5 cmm.	Growth obtained from circ. 5 cmm.
1,000 ,, ,,	—	Growth obtained from circ. 10 cmm.	6 colonies from 15 cmm.	Growth obtained from circ. 5 cmm.	Growth obtained from circ. 5 cmm.
10,000 ,, ,,	—	Growth obtained from circ. 10 cmm.	6 colonies from 15 cmm.	Growth obtained from circ. 5 cmm.	Growth obtained from circ. 5 cmm.
100,000 ,, ,,	40 colonies developed from 10 cmm.	Growth obtained from circ. 10 cmm.	2 colonies from 10 cmm.	Growth obtained from circ. 5 cmm.	Growth obtained from circ. 5 cmm.
1,000,000 ,, ,,	—	Growth obtained from circ. 10 cmm.	0 colonies from 5 cmm.	Growth obtained from circ. 5 cmm.	Growth obtained from circ. 5 cmm.
10,000,000 ,, ,,	0 colonies developed from 20 cmm.	Growth obtained from circ. 10 cmm.	0 colonies from 5 cmm.	Sterile	—

M. G. who had been a martyr to furunculosis, had recently undergone three successive therapeutic inoculations with sterilized staphylococcus cultures.

TABLE IX.—*Exhibiting the Results obtained on adding a Sterilized Culture of Staphylococcus to a Mixture of Serum and Living Typhoid Culture.*

Dilutions in which the Living Typhoid Culture was employed.	Capillary Testing Pipettes were filled in with									
	1 vol. F. N. W.'s Serum, 1 vol. Dilution of Living Typhoid Culture, and		1 vol. A. E. W.'s Serum, 1 vol. Dilution of Living Typhoid Culture, and		1 vol. E. A. S.'s Serum, 1 vol. Dilution of Living Typhoid Culture, and		1 vol. W. G. L.'s Serum, 1 vol. Dilution of Living Typhoid Culture, and		1 vol. W. G. L.'s Serum, 1 vo. Dilution of Living Typhoid Culture containing 6,500,000,000 *T.B.* per c.c., and	
	1 vol. Sterile Broth.	1 vol. Sterilized Staphylococcus Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Staphyloccoccus Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Staphylococcus Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Staphylococcus Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Staphylococcus Culture.
2-fold dilution	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth
5 " "	"	"	"	"	"	"	"	"	"	"
10 " "	"	"	Sterile	Sterile	"	"	"	"	"	"
25 " "	"	"	"	"	"	"	"	"	"	"
50 " "	"	"	"	"	"	"	Sterile	"	"	"
100 " "	Sterile	Sterile	"	"	Sterile	"	"	"	"	"
1,000 " "	"	"	"	"	"	Sterile	"	Sterile	Sterile	"
10,000 " "	"	"	"	"	"	"	"	"	"	"
100,000 " "	"	"	"	"	"	"	"	"	"	Sterile

E. A. S. was a man in normal health.

TABLE X.—*Exhibiting the Results obtained when a Sterilized Culture of Staphylococcus is added to a Mixture of Serum and Living Cholera Culture.*

Dilutions in which the Living Cholera Culture was employed.	Capillary Testing Pipettes were filled with							
	1 vol. F. N. W.'s Serum, 1 vol. Dilution of Living Cholera Culture, and		1 vol. A. E. W.'s Serum, 1 vol. Dilution of Living Cholera Culture, and		1 vol. W. G. L.'s Serum, 1 vol. Dilution of Living Cholera Culture containing 2,600,000 Cholera Vibrios per c.c., and		1 vol. W. B. L.'s Serum, 1 vol. Dilution of Living Cholera Culture containing 21,500,000 Cholera Vibrios per c.c., and	
	1 vol. Sterile Broth.	1 vol. Sterilized Staphylococcus Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Staphylococcus Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Staphylococcus Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Staphylococcus Culture.
Undiluted	—	—	—	—	Growth	Growth	Growth	Growth
2-fold dilution . .	Growth	Growth	Growth	Growth	”	”	”	”
5 ” ” . .	”	”	”	”	”	”	”	”
10 ” ” . .	Sterile	Sterile	”	”	”	”	”	”
25 ” ” . .	”	”	Sterile	Sterile	”	”	Sterile	Sterile
50 ” ” . .	”	”	”	”	”	”	”	”
100 ” ” . .	”	”	”	”	”	”	”	”
1,000 ” ” . .	”	”	”	”	Sterile	Sterile	”	”
10,000 ” ” . .	”	”	”	”	”	”	”	”
100,000 ” ” . .	—	—	—	—	”	”	”	”

The method of experimentation adopted was as follows:—Tw series of progressive dilutions of the culture were made, the diluent employed being in the one case sterile nutrient broth, and in the othe case human serum.

A series of equal volumes of each dilution was measured off int capillary testing pipettes. These measured volumes were in the cas of the broth dilutions immediately transferred to the surface of th nutrient agar with a view to the enumeration of the contained staphy lococcus. The serum dilutions, on the contrary, were before implantatio upon agar digested for twenty-four hours at 37° C. with a view t allowing the serum to exert its full effect upon the micro-organisms.

The results are set forth in Table VIII.

An arithmetical calculation based upon the data set forth in Table VII indicates that in the first experiment 10 cmm. of practically undilute serum failed to kill 0·4, and in the second experiment the same quan tity of practically undiluted serum failed to kill 3 of the staphy locoocus employed.

From the fact that the serum does not exert any bactericidal effec upon the staphylococci, we surmised that no bactericidal substance would be extracted from the serum *in vitro* by the addition of a sterilize culture of staphyloccus.

The substantial correctness of this inference was tested by mean of the experiment set forth in Tables IX and X. It must be note that in these experiments we employed, not as in the experiments se forth in Tables I, III and IV, a sterilized broth culture, but a ver dense bacterial suspension made from one or more agar cultures.

It will be seen that with the exception of experiments 3, 4 and 5 i Table IX, where the difference is in each case a very small one, the bacter icidal effect exerted was in no case less in the case of the serum which ha received an addition of sterilized staphylococcus culture than in the cas of the serum which had received only an addition of sterile nutrien broth.

On reviewing the results obtained, we cannot fail to be struck wit the sharp contrast between those obtained with the staphylococcus an those obtained with the typhoid bacillus and cholera vibrio.

We have seen (*a*) that the typhoid bacillus and the cholera vibrio ar killed off in very large numbers by the normal serum.

(*b*) That sterilized cultures of these micro-organisms when adde to the serum *in vitro* extract from this last a bactericidal element.

(*c*) That the introduction of sterilized cultures of these bacteri into the human and animal organism confers upon the animal an increase bactericidal power, with respect to the particular species of micro organisms inoculated.

On the other hand, we have seen in the case of the staphylococcus

(*a*) That this micro-organism is favourably, rather than unfavourably affected by a contact with the normal serum.

TABLE XI.—*Exhibiting the Results obtained by cultivating Mixtures of one volume of a Graduated Dilution of a culture of the Bacillus of Plague and one volume of Broth or of Serum.*

Dilutions in which the Living Plague Culture was employed.	Capillary Testing Pipettes were filled with		
	1 vol. Dilution of Living Plague Culture (cultivated 4 days at 37° C.) and		
	1 vol. Sterile Broth.	1 vol. F. N. W.'s Serum.	1 vol. A. E. W.'s Serum.
10-fold dilution . . .	Growth	Growth	Growth
25 " " . . .	"	"	"
50 " " . . .	"	"	"
100 " " . . .	"	"	"
1,000 " " . . .	"	"	"
10,000 " " . . .	Sterile	"	"
100,000 " " . . .	"	Sterile	"

A. E. W. had undergone an inoculation with "half-a-dose" of Haffkine's plague vaccine two years previously.

TABLE XII.—*Exhibiting the Results obtained by making Graduated Dilutions of a 2 day old Living Plague Culture, with Sterile Broth and Serum respectively; and incubating one, or in most cases two, 10 cmm. volumes of each Dilution after transference to the surface of Nutrient Agar. This transference was in case of the Serum Dilution postponed for 24 hours. The Capillary Testing Pipettes were in the interval kept at a Temperature of 37° C.*

Dilutions of the Living Plague Culture which were employed.	Number of Colonies which developed on the Nutrient Agar in case of the			
	Dilution made with Sterile Broth.	Dilution made with F. N. W.'s Serum.	Dilution made with A. E. W.'s Serum.	Dilution made with E. A. S.'s Serum.
1,000-fold dilution . .	60	Innumerable	Innumerable	Innumerable
10,000 " " (a) .	12	"	"	"
10,000 " " (b) .	14	"	"	"
100,000 " " (a) .	0	60	40	"
100,000 " " (b) .	2	50	35	35
1,000,000 " " (a) .	1	1	1	50
1,000,000 " " (b) .	1	4	—	50

With regard to A. E. W., see note to Table XI.

(*b*) That sterilized cultures of this micro-organism added to the um *in vitro* do not, unless possibly to a very small extent, diminish bactericidal action upon the typhoid bacillus and the cholera vibrio.

Lastly, it would seem from the experiment in the last column of ble VIII, and from certain other observations which will be discussed ewhere :

(*c*) That the introduction of sterilized cultures of the staphylococcus o the human organism does not confer upon the serum any bactericidal wer.

In view of the important bearing of facts such as those just disclosed connexion with the theory of immunity and in connexion with pro- tive inoculation, we now proceeded to draw within the scope of our uiry, on the one hand, the bacillus pestis, and on the other hand, the crococcus Melitensis.

. Data with regard to the Bactericidal power of the Blood as affecting the Bacillus pestis.

The observations recorded below suggest that, as in the case of the phylococcus, a favourable rather than an unfavourable influence is erted upon the plague bacillus by human serum when mixed in equal umes with a plague culture (see Table XI).

The effect of the serum was further investigated by comparing the mber of living plague colonies obtained from equal volumes of progressive utions of a plague culture made (*a*) with sterile nutrient broth, and (*b*) h human serum.

The results obtained are set forth in Table XII.

It will be manifest that the results bear testimony to the absence of a tericidal effect and to a multiplication of the plague bacilli in almost all serum tubes.

Following out the plan pursued in the case of the other micro-organisms ated of above, we now sought to determine whether any bactericidal ment was extracted when a sterilized plague culture was added to a xture of serum and living typhoid or living cholera culture. The thod of investigation was the same as in the staphylococcus experiments bles IX and X), a very dense bacterial suspension being made m one or more agar cultures. The results obtained are given in Tables II and XIV.

It will be seen that the bactericidal power was practically unaffected the addition of a sterilized plague culture.

Data with regard to the Bactericidal Power of the Blood and the Micrococcus Melitensis.

The data obtained in the case of the Malta fever micrococcus hardly m to require anything in the way of verbal comment. They are joined in the form of Tables XV to XIX inclusive.

TABLE XIII.—*Exhibiting the Results obtained on adding a Sterilized Culture of the Plague Bacillus to a Mixture of Serum and Living Culture of Typhoid.*

Dilutions of the Living Typhoid Culture which were employed.	Capillary Testing Pipettes were filled in with							
	1 vol. F. N. W.'s Serum, 1 vol. of a Living Typhoid Culture containing 3,000,000,000 *T.B.* per c.c., and		1 vol. A. E. W.'s Serum, 1 vol. of a Living Typhoid Culture containing 3,000,000,000 *T.B.* per c.c., and		1 vol. F. N. W.'s Serum, 1 vol. of a Living Typhoid Culture, and		1 vol. A. E. W.'s Serum, 1 vol. of a Living Typhoid Culture containing 1,100,000,000 *T.B.* per c.c., and	
	1 vol. Sterile Broth.	1 vol. Sterilized Plague Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Plague Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Plague Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Plague Culture.
Undiluted culture	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth
2-fold dilution	"	"	"	"	"	"	"	"
5 " " . .	"	"	"	"	"	"	"	"
10 " " . .	"	"	"	"	"	"	Sterile	"
25 " " . .	"	"	"	"	"	"	"	"
50 " " . .	"	"	"	Sterile[1]	Sterile[1]	"	"	"
100 " " . .	"	"	"	Growth	Growth	"	"	"
1,000 " " . .	"	"	Sterile	Sterile	Sterile	"	"	"
10,000 " " . .	Sterile	Sterile	"	"	"	Sterile	"	Sterile
100,000 " " . .	"	"	"	"	"	"	"	"

[1] The irregularity was probably due to an accidental overheating of the tube.

TABLE XIV.—*Exhibiting the Results obtained by the Addition of a Sterilized Plague Culture to a Mixture of Serum and Living Culture of Cholera.*

Dilutions in which the Living Culture of Cholera was employed.	Capillary Testing Pipettes were filled in with							
	1 vol. F. N. W.'s Serum, 1 vol. Living Cholera Culture containing 18,000,000, Cholera Vibrios per c.c., and		1 vol. A. E. W.'s Serum, 1 vol. Living Cholera Culture containing 18,000,000 Cholera Vibrios per c.c., and		1 vol. F. N. W.'s Serum, 1 vol. Living Cholera. Culture containing 44,000,000 Cholera Vibrios per c.c., and		1 vol. A. E. W.'s Serum, 1 vol. Living Cholera Culture containing 44,000,000 Cholera Vibrios per c.c., and	
	1 vol. Sterile Broth.	1 vol. Sterilized Plague Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Plague Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Plague Culture.	1 vol. Sterile Broth.	1 vol. Sterilized Plague Culture.
Undiluted culture . . .	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth
2-fold dilution . . .	Sterile	"	Sterile	Sterile	"	"	"	"
5 " " . . .	"	Sterile	"	"	"	"	"	"
10 " " . . .	"	"	"	"	"	"	Sterile	"
25 " " . . .	"	"	"	"	"	"	"	Sterile
50 " " . . .	"	"	"	"	"	"	"	"
100 " " . . .	"	"	"	"	Sterile	Sterile	"	"
1,000 " " . . .	"	"	"	"	"	"	"	"
10,000 " " . . .	"	"	"	"	"	"	"	"

TABLE XV.—*Exhibiting the Results obtained on cultivating equal volumes of Serum and Diluted Micrococcus Melitensis Culture which had remained in contact at 37° C. for 24 hours.*

Dilutions in which the Living Malta Fever Culture was employed	Capillary Testing Pipettes were filled in with — 1 vol. of a 4-day-old Broth Culture of Micrococcus Melitensis, and				
	1 vol. Sterile Broth.	1 vol. F. N. W.'s Serum.	1 vol. W. B. L.'s Serum.	1 vol. A. B.'s Serum.	1 vol. J. W.'s Serum.
10-fold dilution .	Growth	Growth	Growth	Growth	Growth
25 ” ” .	”	”	”	”	”
50 ” ” .	”	”	”	”	”
100 ” ” .	”	”	”	”	”
1,000 ” ” .	”	”	”	”	”
10,000 ” ” .	”	”	”	”	”
100,000 ” ” .	”	”	”	”	”
1,000,000 ” ” .	Sterile	Sterile	Sterile	Sterile	Sterile

TABLE XVI.—*Exhibiting the Results obtained by cultivating at 25° C. in Capillary Tubes equal Volumes of Serum and Diluted Gelatine Culture of the Micrococcus Melitensis.*

Dilutions of the Gelatine Culture which were employed.	Capillary Tubes were filled in with 1 volume of the Culture of Micrococcus Melitensis diluted with Nutrient Gelatine (15% Gelatine) and		
	1 vol. Sterile Broth.	1 vol. F. N. W.'s Serum.	1 vol. A. E. W.'s Serum.
	No. of Colonies which developed in the Tubes.	No. of Colonies which developed in the Tubes.	No. of Colonies which developed in the Tubes.
10-fold dilution . . .	Innumerable	Innumerable	Innumerable
100 " " . . .	"	"	"
1,000 " " . . .	100 (circ.)	100 (circ.)	100 (circ.)
10,000 " " . . .	17	20	14
100,000 " " . . .	15	23	20
1,000,000 " " . . .	3	6	4

The capillary tubes were filled in and the colonies in them were counted under the microscope by the technique described by one of us in the *Lancet*, December 1, 1900, pp. 1556–1560.

TABLE XVII.—*Exhibiting the Results obtained by diluting a 4 day old Culture of the Micrococcus Melitensis with Sterile Broth and Human Serum respectively, and by cultivating 10 cmm., or, where specified, 5 cmm., of each Dilution, on Nutrient Agar. The transference to Nutrient Agar was in the case of the Serum Dilutions postponed for 24 hours. During this interval the Capillary Testing Pipettes were kept at 37° C.*

Dilutions in which the Culture was employed.	Number of Colonies which developed in the case of the Dilutions of Culture No. 1 made with Sterile Broth.	Dilutions of Culture No. 1 made with F. N. W.'s Serum	Dilutions of Culture No. 2 made with Sterile Broth.	Dilutions of Culture No. 2 made with A. E. W.'s Serum.
100-fold dilution	—	Innumerable	—	—
1,000 " "	—	"	—	—
10,000 " " (a)	Innumerable	"	—	Innumerable
10,000 " " (b)	—	"	—	"
100,000 " " (a)	Innumerable	"	113	100 (circ.)
100,000 " " (b)	—	"	68	—
1,000,000 " " (a)	50	"	15	50 (5 cmm.)
1,000,000 " " (b)	—	"	1	—
10,000,000 " "	1	80	—	—

TABLE XVIII.—*Exhibiting the Results obtained by the addition of a Sterilized Dense Suspension of the Micrococcus Melitensis to a Mixture of Serum and Living Typhoid Culture.*

Dilutions in which the Living Typhoid Culture was employed	Capillary Testing Pipettes were filled in with									
	1 vol. Living Typhoid Culture, 1 vol. F. N. W.'s Serum, and		1 vol. Living Typhoid Culture, 1 vol. E. A. S.'s Serum, and		1 vol. Living Typhoid Culture, 1 vol. W. B. L.'s Serum, and		1 vol. Living Typhoid Culture, 1 vol. F. N. W.'s Serum, and		1 vol. Living Typhoid Culture, 1 vol. A. E. W.'s Serum, and	
	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.
Undiluted culture . .	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth
2-fold dilution .	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
5 ,, ,, .	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
10 ,, ,, .	Sterile	,,	,,	,,	,,	,,	,,	,,	,,	,,
25 ,, ,, .	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
50 ,, ,, .	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
100 ,, ,, .	,,	,,	Sterile	,,	,,	,,	,,	,,	,,	,,
1,000 ,, ,, .	,,	,,	,,	,,	Sterile	,,	,,	Sterile	Sterile	Sterile
10,000 ,, ,, .	,,	Sterile	,,	Sterile	,,	Sterile	Sterile	,,	,,	,,
100,000 ,, ,, .	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,

TABLE XIX.—*Exhibiting the Results obtained by the Addition of a Sterilized Dense Suspension of the Micrococcus Melitensis to a Mixture of Serum and Living Cholera Culture.*

Dilutions in which Living Cholera Culture was employed.	Capillary Testing Pipettes were filled with									
	1 vol. Living Cholera Culture, 1 vol. E. A. S.'s Serum, and		1 vol. Living Cholera Culture containing 18,000,000 C. vibrios per c.c. 1 vol. F. N. W.'s Serum and		1 vol. Living Cholera Culture containing 18,000,000 C. Vibrios per c.c., 1 vol. A. E. W.'s Serum, and		1 vol. Living Cholera Culture containing 44,000,000 C. Vibrios per c.c., 1 vol. F. N. W.'s Serum, and		1 vol. Living Cholera Culture containing 44,000,000 C. Vibrios per c.c., 1 vol. A. E. W.'s Serum, and	
	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.	1 vol. Sterile Broth.	1 vol. Sterilized M.M. Culture.
Undiluted culture . .	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth	Growth
2-fold dilution .	"	"	Sterile	"	Sterile	Sterile	"	"	"	"
5 " " .	"	"	"	"	"	"	"	"	"	"
10 " " .	"	"	"	"	"	"	"	"	Sterile	Sterile
25 " " .	"	"	"	Sterile	"	"	"	"	"	"
50 " " .	Sterile	Sterile	"	"	"	"	"	Sterile	"	"
100 " " .	"	"	"	"	"	"	Sterile	"	"	"
1,000 " " .	"	"	"	"	"	"	"	"	"	"
10 000 " " .	"	"	"	"	"	"	"	"	"	"
100,000 " " .	"	"	"	"	"	"	"	"	"	"

Tables XV and XVI show that human serum, when mixed volume volume with cultures of micrococcus Melitensis, is without action upon micro-organism.

Table XVII establishes that even the undiluted serum is entirely iout bactericidal action, and that a multiplication of the micro-organism y take place in this medium.

Tables XVIII and XIX establish that the anti-bactericidal effect rted by the addition of a dense suspension of micrococcus Melitensis n human serum is quite insignificant.

Conclusions.

On reviewing the experimental data which we have set forth, it would n clear that—

1. Human serum exerts a powerful bactericidal effect upon the typhoid illus and the cholera vibrio, while it is without bactericidal action n the staphylococcus pyogenes, B. pestis, micrococcus Melitensis, l, so far as we have gone) upon the streptococcus pyogenes and diphtheriae.

2. Sterilized cultures of those species of pathogenic micro-organisms ch are killed by the serum, appear, in contra-distinction to those cies of micro-organisms which are not affected by the serum, to possess power of directly abstracting a bactericidal element from the blood.

The first of these generalizations appears to possess a far-reaching ificance in connexion with the general theory of immunity.

(*a*) It has an obvious bearing on the question of the mechanism by ich bacteria are destroyed in the organism.

(*b*) It also bears on the question as to whether the bactericidal ion is acquired only after withdrawal from the organism, and after the ntegration of leucocytes.

For it would seem difficult to assume that the bactericidal power the serum is only a particular manifestation of a digestive power ginally resident in the leucocyte, when we have realized that the serum rts a bactericidal action only on particular species of micro-organisms, ile the leucocyte exerts a digestive action on bacteria generally.

The second of the generalizations arrived at above would seem to nt to the bactericidal effects being the result of definite chemical nbinations occurring between the bactericidal substance or substances the blood and the affected bacteria.

In conclusion, reference may be made to a possible relation between danger or relative absence of danger associated with the hypodermic culation of different species of bacteria, and the effect or absence of ect of the blood upon these micro-organisms. A notable contrast tains in this respect between the event of inoculations of cholera and hoid on the one hand, and plague and Malta fever on the other hand.

While inoculation with living cultures of cholera is, as has been

shown in connexion with Haffkine's anticholera inoculations, practical unassociated with risk, and while inoculations with small quantiti of living typhoid bacilli are—judging from the event of an experiment inoculation undertaken by one of us, and from the immunity from accide which has attended wholesale manipulations with this micro-organis —associated with only slight risk, the results are quite other in t case of even minimal inoculations of plague and Malta fever culture

That extreme risk attaches to the inoculation of even minimal qua tities of living plague bacilli is attested by the numerous cases of plag which have supervened upon the accidental inoculation of infect material into small superficial scratches.

The risk attaching to even minimal inoculations of the micrococc Melitensis is less well known. Six cases of the disease have occurred connexion with bacteriological work on Malta fever undertaken Netley, and two further cases have originated at the Royal Nav Hospital, Haslar, and in the Philippines respectively, in connexion wit bacteriological work.

Of the cases ocurring at Netley, one originated from an accidental pric with a needle of a syringe containing a Malta fever culture ; a secon arose in connexion with an experimental inoculation ; and a third h recently occurred in connexion with the accidental projection of the er of a contaminated capillary sedimentation tube into the eye. The thre other cases at Netley arose apart from a recognized inoculation in t case of observers working with living cultures. It would seem difficu to conceive of inoculations with quite minimal quantities of cultur being so effectual in the case of micro-organisms subject to the bactericid action of the blood and lymph.

On the Comparative Bactericidal Effect of Human Blood drawn off and tested under Aerobic and Anaerobic Conditions.[1]

By A. E. Wright.

From the Laboratory of the Pathological Department, Army Medical School, Netley.

v view of the fundamental theoretical importance which attaches to ıe assumption that the bactericidal power of the blood is acquired only ter withdrawal from the organism, and, in particular, after the disin-gration of the leucocytes under the influence of air and contact with ıe wall of unoiled or unparaffined receptacles, it seemed important re-investigate the question ; I have therefore endeavoured to ascertain hether there is any constant and important difference between the ıcteridical power of human blood (*a*) drawn off and tested by the ɜrobic procedure described in Section I[2] of this paper, and (*b*) drawn f and tested by the anaerobic procedure described in Section II[2] of ıis paper.

The results of this investigation are set forth below in tabular form, ıd it will be observed that while they are, of course, inconclusive on ıe wider question of the derivation of the bactericidal substances of ıe serum, they would seem definitely to show that neither contact with ıe external air, nor contact with ordinary glass surfaces, exerts any ıportant influence on the bactericidal power exerted by human blood ɔon the typhoid bacillus and the cholera vibrio.

[1] Reprinted from the *Proceedings of the Royal Society*, vol. lxxi, 1902.
[2] The portions of the paper which deal with the technique are not reprinted here.

Table exhibiting the Bactericidal Effect produced by one and the same Serum (a) drawn off and tested by the Aerobic Procedure described in Section I; and (b) drawn off and tested by the Anaerobic Procedure described in Section II.

Dilutions in which the Culture was employed.	Capillary Testing Pipettes were filled in with—																			
	1 vol. A. E. W.'s Serum, 1 vol. of a Typhoid Culture, containing 156,000,000 T. B. per c.c.		1 vol. W. B. L.'s Serum, 1 vol. of a Typhoid Culture, containing 156,000,000 T. B. per c.c.		1 vol. F. N. W.'s Serum, 1 vol. of a Typhoid Culture containing 156,000,000 T. B. per c.c.		1 vol. A. E. W.'s Serum, 1 vol. of a Typhoid Culture containing 120,000,000 T. B. per c.c.		1 vol. F. N. W.'s Serum, 1 vol. of a Typhoid Culture containing 150,000,000 T. B. per c.c.		1 vol. A. E. W.'s Serum, 1 vol. of a Typhoid Culture containing 100,000,000 T. B. per c.c.		1 vol. A. B.'s Serum, 1 vol. of a Typhoid Culture containing 220,000,000 T. B. per c.c.		1 vol. J. N.'s Serum, 1 vol. of a Typhoid Culture containing 540,000,000 T. B. per c.c.		1 vol. A. E. W.'s Serum, 1 vol. of a Cholera Culture containing 18,000,000 Cholera Vibrios per c.c.		1 vol. A. E. W.'s Serum, 1 vol. of a Cholera Culture containing 60,000,000 Cholera Vibrios per c.c.	
	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.	Aerobic Procedure.	Anaerobic Procedure.
Undiluted . . .	—	—	—	—	—	—	—	—	Growth	Growth	—	—	—	—	—	—	Growth	Growth	Growth	Growth
2-fold dilution	—	—	—	—	—	—	—	—	,,	,,	Growth	Growth	—	—	Growth	Growth	Sterile	Sterile	,,	,,
5 ,, ,,	Sterile	Sterile	Growth	Sterile	Growth	Growth	Growth	Growth	,,	,,	,,	,,	—	—	Sterile	,,	,,	,,	,,	,,
10 ,, ,,	,,	,,	Sterile	,,	,,	,,	,,	,,	,,	,,	Sterile	,,	Growth	Growth	,,	,,	,,	,,	,,	,,
25 ,, ,,	,,	,,	,,	,,	,,	Sterile	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
50 ,, ,,	,,	,,	,,	,,	,,	,,	Sterile	Sterile	Sterile	Sterile	,,	,,	Sterile	Sterile	,,	,,	,,	,,	Sterile	Sterile
100 ,, ,,	,,	,,	,,	,,	,,	,,	Growth	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
1,000 ,, ,,	,,	,,	,,	,,	,,	,,	Sterile	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
10,000 ,, ,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,
100,000 ,, ,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,	,,

A. E. W. had been inoculated against typhoid; W. B. L., F. N. W., and A. B. were normal men; J. N. had recently convalesced from typhoid. The sera were in each case tested within 2–3 hours after the blood had been withdrawn. The cultures were in all cases aerobically grown 24-hour old broth cultures. The serum was in each case allowed to act upon the culture for 18–24 hours at a temperature of 37° C.

An Experimental Investigation of the Role of the Blood Fluids in connexion with Phagocytosis.[1]

By A. E. Wright and Stewart R. Douglas.

From the Laboratory of the Pathological Department, St. Mary's Hospital, London, W.

Methods of Experimentation.—Accuracy of the Method, and Special Points which come up for consideration in connexion with it—Does the Substitution of another Medium for the (Citrated) Blood Plasma which Bathes the Corpuscles exert an Influence on Phagocytosis?—Do the Blood Fluids co-operate in Phagocytosis by exerting a Direct Stimulating Effect upon the Phagocytes, or by effecting a Modification in the Bacteria?—Does the Unheated Serum contain, in addition to Elements which render the Bacteria more liable to Phagocytosis (Opsōnins), also Elements which directly stimulate the Phagocytes (Stimulins)?

It is still a matter of uncertainty whether the blood fluids perform any rôle in connexion with phagocytosis.

Certain facts suggest that the rôle of the blood fluids, if it comes into consideration at all, is very subordinate. The facts we have in view are, on the one hand, the facts brought forward by Metchnikoff to show that bacteria may be ingested in the living condition, and on the other hand those brought forward by one of us in conjunction with Captain F. Windsor, I.M.S.,[2] which show that the human serum exerts absolutely no bactericidal action on the staphylococcus pyogenes, the micrococcus Melitensis and the plague bacillus.

These facts are, however, not conclusive. They are not inconsistent with the idea that the blood fluids, apart from actually killing the particular pathogenic bacteria here in question, may in some way co-operate in their destruction.

What are required for the resolution of the problem are experiments in which the phagocytes are tested apart from the blood fluids.

The experimental methods which we now pass on to describe enable these crucial experiments to be made.

[1] Reprinted from the *Proceedings of the Royal Society*, Vol. lxxii, 1903.

[2] Wright and Windsor, *Journal of Hygiene*, vol. ii, No. 4, Oct., 1902 (pp. 1–72 *supra*).

Method of Experimentation.

We have employed a modification of the method of measuring the phagocytic power of the blood, which was devised by Major W. B. Leishman, R.A.M.C., at that time our fellow-worker.[1]

In the procedure described by this author equal volumes of a bacterial suspension of appropriate density and of blood drawn from the finger are measured off in a capillary tube, mixed on a slide and covered in with a cover-glass. The blood and bacterial culture are then left in contact for fifteen minutes in an incubator standing at blood heat. After this interval the cover-glass is, if necessary, loosened from the side by a drop of physiological salt solution, and the slide and cover-glass are drawn apart by a sliding movement.

The films thus obtained are stained by Leishman's[2] modification of Romanovski's stain, and are subjected to examination under an immersion lens. By enumerating the bacteria ingested in a number of polynuclear white blood corpuscles and dividing, an average is obtained. This average is taken as the measure of the phagocytic power of the blood. It is compared, when comparative experiments are made, with the phagocytic power of a normal blood.

We have modified this method for our purposes (*a*) by conducting the phagocytosis in capillary tubes, making afterwards film preparations in the ordinary way; (*b*) by decalcifying the blood with citrate of soda, thus avoiding the complications introduced by blood coagulation, and making it possible to separate the white corpuscles from the blood fluids by centrifugalization, decantation and washing.

Three different procedures, varying only in details, were employed in our experiments.

Procedure No. 1, *employed where nothing more than a Comparison between Bloods from different Sources or Blood subjected to Different Conditions is required.*

Having provided ourselves with a simple capillary pipette, furnished with a rubber teat and a pencil mark on the stem, we aspirate into the stem of the pipette—dividing off by bubbles of air in accordance with the procedure introduced by one of us—one volume of blood from the finger, one volume of a 1 per cent. solution of citrate of soda in physiological salt solution, and one volume of a bacterial[3] suspension made by shaking up a 24-hour agar culture in physiological salt solution, and centrifugalizing so as to remove any bacterial clumps. We mix together the three equal

[1] *British Medical Journal*, Jan. 11, 1902.
[2] *British Medical Journal*, 1901.
[3] This bacterial suspension may conveniently contain about 10,000,000,000 bacteria in the cubic centimetre. The number may be readily adjusted by the help of the method of enumeration under the microscope described by one of us in the *Lancet*, July 5, 1902.

olumes of blood, bacterial suspension and citrate of soda solution, by lowing these out upon a clean slide and re-aspirating several times in uccession. Mixture completed, an aliquot portion of the mixed fluids, uch as suffices for our purposes, is drawn up into the capillary stem, and he orifice of the capillary tube is sealed in the flame. This done, the ipette is placed either in an incubator standing at 37° C., or in a vessel f water kept at this temperature.

After the lapse of fifteen minutes we break off the extremity of the ipette, *carefully mix* the contents so as to get an average sample, and roceed to make films, and then to stain them by Leishman's dye.

rocedure No. 2, where we desire to elicit separately the rôle of the White Corpuscles and the Blood Fluids in Phagocytosis, and to study the Effect produced by Experimental Modification of one or other of these Elements separately.

Having provided ourselves with a capsule with a recurved limb (such capsule has already been figured in a previous communication),[1] we ntroduce into it such a quantity of mercury as will fill it to about one-hird of its capacity. Having marked off by a pencil mark (made with a ass writing pencil) the level at which the upper surface of the mercury ands, we displace the mercury in such a manner as to cause it to occupy he middle instead of the lower region of the capsule. We again mark off n the outside of the capsule the upper limit of the mercury.

Then, emptying out this last, we bend round in the flame the curved mb in such a manner as to cause it to lie in the plane of the equator f the capsule. This enables us to siphon into it from a watch glass, lled and placed ready to hand, the citrate of soda solution. We introduce f this solution such a quantity as suffices to fill the capsule up to the vel of the first pencil mark. This done, we draw blood from the finger nd let it run into the capsule until the combined volume of citrate of oda solution and blood attains the level of the second pencil mark.

Having sealed up the upper orifice in the flame—rarefying as we o so the air in the interior of the capsule by the application of warmth—e shake up the contents and suspend the capsule by means of its curved mb into the receptacle of the hand centrifugal machine.

When centrifugalization has caused the corpuscles to settle to the ottom, we pipette off and reserve the supernatant citrated plasma nd replace it by physiological salt solution. In conducting this last peration we employ a capillary pipette, and we carry down its orifice to he very bottom of the capsule in such a manner as effectively to mix up he corpuscles and the newly added fluid. We wash and centrifugalize n this manner three times. The upper layers of the corpuscular deposit,

[1] Wright, *Roy. Soc. Proc.*, vol. lxxi, 1902.

containing as they do a large proportion of white corpuscles, supply th
phagocytes required for experimentation.

In the experiments set forth below we mixed in each case three volume of the upper layers of the washed corpuscular deposit with three volume of blood fluid and one volume of a staphylococcus suspension, containin generally from 7,000–10,000 million staphylococci per c.c. The mixtu of corpuscles, blood fluid and staphylococci was kept in each case fo fifteen minutes at a temperature of 37° C., in order to give opportunit for the occurrence of phagocytosis.

Procedure No. 3, *employed where we desire to obtain Citrated Serum fo Comparison with the Citrated Plasma furnished by Procedure No.* 2.

Where we desire to obtain citrated serum for comparison with th citrated plasma furnished by Procedure No. 2, we graduate our bloo capsule in precisely the same manner. Having filled in with blood fro the finger up to the first mark, we allow it to clot, and we then introdu into the capsule from a capillary pipette a sufficiency of citrate of sod solution to complete up to the second mark. Finally, we churn up th citrate of soda solution with the blood clot and then centrifugalize.

Accuracy of the Method, and Special Points which come up for Considera tion in connexion with it.

The accuracy of the method is attested by the concordant results se forth below of the large number of experiments which we conducted i duplicate. We desire to point out that the results incorporated belo represent not exceptional fortunate achievements, but simply what ma be obtained by the ordinary every-day application of the method.

Before dismissing the consideration of the experimental method, i may be well to elucidate very briefly three points which suggest themselve for consideration in connexion with it.

The first of these relates to the calibre of the capillary tubes.

In our earlier experiments we considered it advisable, with a view t providing against a possible cause of fallacy, to conduct our experiment in capillary tubes of a standard calibre. The tubes were in each cas calibrated by the method described by one of us,[1] to wit, by introducin into the wide end of a tube drawn out in the flame 5 cmm. of mercur from an "automatic pipette," and marking off that portion of the capil lary stem where this quantum of mercury formed a column 5 cm. in length The experiments which we conducted with calibrated tubes are thos which occupy the two next following sections of this paper.

[1] *Transactions of the Roy. Medico-Chirurg. Soc.*, vol. lxxxiv and *Lancet*, Jul 5, and Dec., 1902.

In our later experiments, to wit, in the experiments which occupy the ter sections of this paper, we discarded calibrated for uncalibrated tubes, aking only the condition that the capillary tubes employed in comparave experiments should appear to the eye to be more or less comparable . calibre. It will be seen, on looking into our results, that the concordice obtained was not less in the case where uncalibrated tubes were nployed than in the case where calibrated tubes were employed.

Different results, however, emerge when experiments in duplicate are onducted with tubes presenting extreme differences in calibre. In a ries of comparative experiments, in which we employed in each case an most hair-fine tube as a fellow to a tube almost too large to be reckoned s a capillary tube, the results were irregular, being generally but not onsistently in favour of the narrower tube.

A *second point* which comes up for consideration is the possible effect the addition of citrate of soda to the blood.

The concentration of the solution in particular comes into consideraon. Finding that phagocytosis is inhibited when the white corpuscles e bathed in a medium containing 3 per cent. of citrate of soda, we took ie precaution to add to the blood in comparative experiments presely the same amounts of citrate of soda. It may be noted that the iorphological structure of the white corpuscles is extremely well prerved, and phagocytosis proceeds actively in a medium containing up to ·5 per cent. of citrate of soda.

The *third* and last *point* to be considered relates to the maintenance of ie activity of the phagocytes for a sufficient period after they have been ithdrawn from the organism and have been subjected to the procedures escribed above. It will be manifest that, apart from a maintenance of ie activity of the phagocytes under the conditions which come into conderation here, it would be impracticable to compare the results of xperiments instituted in succession with one and the same quantum of ashed corpuscles, or to compare the phagocytic power of different bloods, nless in the case where these were withdrawn from the organism multaneously.

A number of experiments undertaken with a view of obtaining inforation with regard to the point here raised have shown us that the phagoytic power is well maintained under the circumstances of our experiments. ven after the lapse of three days (our observations have not extended eyond this limit) the phagocytic power has not declined to less than onealf or one-third of that of the blood freshly drawn. We have found o indication of a variation within the space of a few hours.

These preliminary points having been dealt with, we may pass to the onsideration of the problem to which attention was directed in the pening paragraph of this paper.

Does the Substitution of another Medium for the (Citrated) Blood Plasm which bathes the Corpuscles exert an Influence on Phagocytosis ?

1. *Comparative Experiments with Citrated Plasma and Citrated Serun (obtained respectively as described in Connexion with Procedure and 3).*

Experiment 1.

A.

S. R. D.'s *plasma*, 3 vols. ; staphylococcus suspension, 1 vol. ; A. E. W.'s cor puscles, 3 vols.

Tube 1.—Phagocytic count (obtained by averaging the number of staphylococci ingested by 20 P.W.B.C.) . . 34·6
Tube 2.— Do. do. 33·6 } **34·1**

B.

S. R. D.'s *serum*, 3 vols. ; staphylococcus suspension, 1 vol. ; A. E. W.'s cor puscles, 3 vols.

Tube 1.—Phagocytic count (obtained as above) . . . 35·6
Tube 2.— Do. do. . . . 33·8 } **34·7**

Experiment 2.

A.

A. E. W.'s *plasma*, 3 vols. ; staphylococcus suspension, 1 vol, ; A. E. W.' corpuscles, 3 vols.

Tube 1.—Phagocytic count (obtained as above) . . . 31·2
Tube 2.— Do. do. . . . 36·0 } **33·6**

B.

A. E. W.'s *serum*, 3 vols. ; staphylococcus suspension, 1 vol. ; A. E. W.'s cor puscles, 3 vols.

Tube 1.—Phagocytic count (obtained as above) . . . 31·2
Tube 2.— Do. do. . . . 33·0 } **32·1**

It is clear that the phagocytic power is uninfluenced by the substitutio of serum for plasma.

2. *Comparative Experiments with Ordinary (Uncitrated) Serum Unheate and Heated for* 10–15 *min. to* 60–65° C. *and then cooled.*

Experiment 1.

A.

A. E. W.'s *unheated serum*, 3 vols. ; staphylococcus suspension, 1 vol. ; A. E. W. corpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 20 P.W.B.C. enumerated and averaged) . . . . **17·4**
Tube 2.— Do. do. **19·8**

B.

A. E. W.'s *heated serum*, 3 vols. ; staphylococcus suspension, 1 vol. ; A. E. W.' corpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 52 P.W.B.C. enumerated and averaged) . . . . **0·6**
Tube 2.—Phagocytic count (bacteria in 46 P.W.B.C. enumerated and averaged) . . . . **3·4**

Experiment 2.

A.

S. R. D.'s *unheated serum*, 3 vols. ; staphylococcus suspension, 1 vol. ; S. R. D.'s rpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 20 P.W.B.C. enumerated and averaged) . . . . 18·5
Tube 2.—Phagocytic count (bacteria in 22 P.W.B.C. enumerated and averaged) . . . . 16·0

B.

S. R. D.'s *heated serum*, 3 vols. ; staphylococcus suspension, 1 vol. ; S. R. D.'s corpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 29 P.W.B.C. counted and averaged) . . . . 1·5
Tube 2.—Phagocytic count (bacteria in 46 P.W.B.C. counted and averaged) . . . . 1·8

Experiment 3.

A.

A. E. W.'s *unheated serum*, 3 vols. ; staphylococcus suspension, 1 vol. ; A. E. W.'s rpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 9 P.W.B.C. counted and averaged). . . . . 25·4
Tube 2.—Phagocytic count (bacteria in 18 P.W.B.C. counted and averaged). . . . . 16·0

B.

A. E. W.'s *heated serum*, 3 vols. ; staphylococcus suspension, 1 vol. ; A. E. W.'s rpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 20 P.W.B.C. counted and averaged) 0
Tube 2.— Do. do. 0

Experiment 4.

A.

S. R. D.'s *unheated serum*, staphylococcus suspension and corpuscles in the me proportions as before.

Phagocytic count (bacteria in 15 P.W.B.C. counted and averaged) 15·7

B.

S. R. D.'s *heated serum*, staphylococcus suspension, and corpuscles in the same oportions as before.

Phagocytic count (bacteria in 45 P.W.B.C. counted and averaged) 0·2

These experiments show that we must ascribe an important rôle to e blood fluids in connexion with phagocytosis.

For the alternative assumption, the supposition, to wit, that inhibiting ements are developed in the serum during the process of heating, is butted by the results of a series of control experiments, which showed at the phagocytes display no greater activity in a medium of physiolo-cal salt solution than in a medium of heated serum.

It is further rebutted by the circumstance that the activity of phagocy-sis falls off at the same rate when the unheated serum is diluted with lt solution as when it is diluted with heated serum.

The experiment whose results are tabulated below illustrates this la
point.

Results of a Comparison made between the Activating Power of (a) *Unheat Serum diluted with Heated Serum, and* (b) *Unheated Serum diluted wi Physiological Salt Solution.*

In each case 3 vols. of serum dilution were mixed with 1 vol. of staphy lococcus suspension and 3 vols. of washed corpuscles.

Dilution in which the unheated serum was employed.	Average phagocytic count obtained in the case where the unheated serum was diluted with previously heated serum.	Average phagocytic cou obtained in the case where th unheated serum was dilute with physiological salt solutio
3-fold	—	34·2
6-fold	27·4	27·2
12-fold	23·1	30·5
24-fold	20·6	24·8
48-fold	5·0	4·95
96-fold	—	0·8
192-fold	—	0·6

It is clear that we may conclude that the heated serum, like the sa solution, acts merely as an inert diluent, and that we may, in referrin to such heated serum, characterize it simply as "inactivated serum. It is further clear that we may legitimately[1] ascribe the small amount o phagocytosis which occurred in Experiments 1, 2, and 4 *supra*, to th presence of a residuum of unheated serum, which the washing operatio had failed to separate from the corpuscles.

Do the Blood Fluids co-operate in Phagocytosis by exerting a Dire Stimulating Effect upon the Phagocytes, or by effecting a Modif cation in the Bacteria ?

The following experiments were instituted with a view to elucidatin the problem as to the nature of the activating influence exercised by th blood fluids. It will be seen that a comparison is in each case institute between serum inactivated (by heating) before it came in contact wit either bacteria or white corpuscles, and serum inactivated after it ha come in contact with the bacteria, but before it had come in contact wit the white corpuscles :—

Experiment 1.

A.

S. R. D.'s *inactivated* serum, 3 vols. ; staphylococcus suspension (previous heated to 60° C. for 15 minutes and cooled), 1 vol. ; S. R. D.'s corpuscles, 3 vol

[1] *At the time this was written the alternative that the residual phagocytosis might due to 'spontaneous phagocytosis' did not suggest itself.*

Tube 1.—Phagocytic count (bacteria in 20 P.W.B.C. counted and averaged) . . . . 3·4
Tube 2.— Do. do. 3·35

B.

. R. D.'s *unheated* serum, 3 vols.; digested at 37° C. for 15 minutes, with 1 of staphylococcus suspension, then heated to 60° C. for 15 minutes and cooled. vols. of the above mixed with 3 vols. of S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 20 P.W.B.C. counted and averaged) . . . . 27·5
Tube 2.— Do. do. 28·9

Experiment 2.

A.

. E. W.'s *inactivated* serum, 3 vols.; staphylococcus suspension, 1 vol.; digested ther for 15 minutes at 37° C., then heated for 10 minutes to 60° C. and cooled. . vols. of the above mixed with 3 vols. of S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 20 P.W.B.C. counted and averaged) . . . . 4·0
Tube 2.— Do. do. 3·2

B.

. E. W.'s *unheated* serum, 3 vols.; staphylococcus suspension, 1 vol.; digested ther for 15 minutes at 37° C., then heated for 10 minutes to 60° C. and cooled. vols. of the above added to 3 vols. of S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 24 P.W.B.C. counted and averaged) 33
Tube 2.—Phagocytic count (bacteria in 19 P.W.B.C. counted and averaged) 36

Experiment 3.

A.

. R. D.'s *inactivated* serum, 3 vols.; staphylococcus suspension (previously ed to 75° C. and cooled), 1 vol.; digested together for 15 minutes at 37° C. vols. of the above added to 3 vols. of S. R. D.'s corpuscles.

Phagocytic count (bacteria in 30 P.W.B.C. counted and averaged) . 4·2

B.

. R. D.'s *unheated* serum, 3 vols.; staphylococcus suspension (previously ed to 75° C. and cooled), 1 vol.; digested together for 15 minutes at 37° C., heated for 10 minutes to 60° C., and cooled. vols. of the above added to 3 vols. of corpuscles.

Tube 1.—Phagocytic count (bacteria in 15 P.W.B.C. counted and averaged) . . . . 28·2
Tube 2.— Do. do. 31·0

Ve have here conclusive proof that the blood fluids modify the bacteria manner which renders them a ready prey to the phagocytes.

Ve may speak of this as an "*opsōnic*" effect (opsōno—*I cater for; I are victuals for*), and we may employ the term "opsōnins" to desig- the elements in the blood fluids which produce this effect.

Does the Unheated Serum contain, in addition to Elements which rend the Bacteria more liable to Phagocytosis (Opsonins), also Elemen which directly stimulate the Phagocytes (Stimulins) ?

We have sought to elucidate this question by three separate methods. In the first series of experiments, we experimented with staphylococ which had been exposed to high temperatures (115° C.) with the desig of rendering them insusceptible to the opsonic power of the blood fluid Our expectations from this method—expectations based on the fact th we had noticed that typhoid bacilli acquired, when heated to over 70° C., resistance to the bacteriolytic effect of the blood fluids—were unrealize We found that the quantitative differences between the phagocytosis heated and unheated serum respectively were not less in the case of staph lococci which had been exposed to a temperature of 115° C., than in th case of staphylococci which had not been subjected to high temperature

In a second series of experiments we substituted for suspensions staphylococci suspensions of particles, which we assumed would uninfluenced by the opsonic power of the blood. The results of the experiments, conducted both with carmine particles and with India ink, were inconclusive by reason of the circumstance that we were n able to obtain any satisfactory enumerations. An impression was, how ever, left on our minds that phagocytosis was in every case more acti in unheated than in the heated serum.

A third method of experimentation was then resorted to. In a fir operation we mixed and digested together at blood heat a suspension staphylococci and unheated serum. After allowing what we suppose would be a sufficient interval for the exhaustion of the effect of the seru upon the bacteria, we divided the mixture into two portions. Whi the first of these portions was mixed with the corpuscles without unde going any further treatment, the other was heated to 60° C., and coole before it was so mixed. In each case the phagocytic power exerted w greater in the case where the heating was omitted, and the differences we not less marked where the serum had been digested with the bacter for fifty minutes and one hour respectively than in the case where it ha been digested with these only for fifteen minutes.

These results are ambiguous.

The question as to whether the blood fluids contain, in addition opsonins, also an element which directly stimulates the phagocytes, r mains for the present unsolved.

The third series of experiments, which has just been adverted to, subjoined :—

Experiment 1.

S. R. D.'s serum, 3 vols. ; staphylococcus suspension, 1 vol. ; digested f 15 minutes at 37° C.

A.

4 vols. of the above mixture heated to 60° C. for 15 minutes, then cooled and ed to 3 vols. of S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 16 P.W.B.C. enumerated and averaged) . . . . 22·0
Tube 2.—Phagocytic count (bacteria in 31 P.W.B.C. enumerated and averaged) . . . . 20·7

B.

4 vols. of the above mixture added directly to 3 vols. of S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 16 P.W.B.C. counted and averaged) 27
Tube 2.—Phagocytic count (bacteria in 28 P.W.B.C. counted and averaged) 28

Experiment 2.

S. R. D.'s unheated serum, 3 vols.; staphylococcus suspension, 1 vol.; digested 50 minutes at 37° C.

A.

4 vols. of the above mixture heated to 60° C. for 20 minutes, then cooled and ed to 3 vols. of S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 20 P.W.B.C. enumerated and averaged) . . . . 13·5
Tube 2.— Do. do. 17·1

B.

4 vols. of the above mixture added directly to 3 vols. S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 15 P.W.B.C. enumerated and averaged) . . . . 40·6
Tube 2.— Do. do. . 44·5

Experiment 3.

S. R. D.'s unheated serum, 3 vols.; 1 vol. of staphylococcus suspension; digested ther 1 hour at 37° C.

A.

4 vols. of the above mixture heated to 60° C. for 10 minutes, cooled and added vols. of S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 16 P.W.B.C. enumerated and averaged) . . . . 15·1

B.

4 vols. of the above mixture added directly to 3 vols of S. R. D.'s corpuscles.

Tube 1.—Phagocytic count (bacteria in 16 P.W.B.C. counted and averaged) . . . . 22·1

In conclusion we would briefly refer to the following points:—

The opsonic power of the blood fluids disappears gradually on standing, n when the serum is kept in a sealed capsule sheltered from the light. After five or six days we have found the opsonic power of the serum t under these conditions to stand at little more than half of what it originally.

The opsonic power of the blood fluids is but little impaired by th action of heat until temperatures above 50° C. are arrived at. Th following are the results of a typical experiment :—Phagocytic cou obtained with the serum before exposure to heat, 12·7 ; with the sam serum heated for ten minutes to 45° C., 13·1 ; with the same serum heate for ten minutes to 50° C., 10·2 ; with the same serum heated for te minutes to 55° C., 5·7.

The opsonic [1] power of the serum is diminished when this last has be digested with typhoid bacteria. This " anti-opsonic " effect may compared with the " anti-bactericidal " effect [2] obtained on digesting th serum with typhoid or cholera cultures.

The opsonic power of the blood fluids is diminished while the phagocyt capacity of the W.B.C. is preserved when the blood fluids and corpuscl are separately digested with Daboia venom. In the anti-opsonic effe exerted by the venom on the blood fluids, we have probably the explan tion of the reduced resistance to septic invasion which supervenes up viper bites.

It would seem probable that the bacteriolytic, bactericidal, and bact rio-opsonic effects exerted by the blood fluids are each in their degr manifestations of a digestive power exerted by the blood fluids bacteria brought into contact with them.

Lastly, a fact which has a practical importance in connexion with t study of immunity may be adverted to. It will be manifest that we ha not exhausted the study of a condition of immunity when we have measur the phagocytic power of the white corpuscles, and the agglutinating, ba teiolytic, and bactericidal power of the blood fluids. We must, in co nexion with these last, take into consideration also the opsonic effect.

A concrete example may be added to show the kind of elucidati which may be gained from an inquiry which takes into considerati also the factor last mentioned.

The condition of immunity to staphylococcus which can be induc in patients unduly susceptible to staphylococcus infections, by the inoc lation of properly adjusted doses of a sterilized staphylococcus cultu is, as was shown by one of us, associated with an increase of the phagocy power [3] and is unaccompanied by any development of a bactericid power in the blood fluids.

The result of the subjoined blood examinations undertaken upon patient who had been subjected to two successive therapeutic inocu tions of a sterilized staphylococcus culture, suggests that the increas phagocytic power may depend upon an increase in the opsonic power the blood fluids.

[1] *We had here in view the staphylo-opsonic power, and we failed to reflect that serum, after digestion with the typhoid bacilli, might exert a toxic effect upon the was leucocytes.*

[2] Wright and Windsor, *Journal of Hygiene*, vol. ii, No. 4, 1902 (pp. 44–72 *sup*

[3] *Lancet*, March 29, 1902 (pp. 99 *et seq.*).

A.

A. E. W.'s serum, 3 vols.; staphylococcus suspension, 1 vol.; and A. E. W.'s hed corpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 20 P.W.B.C. counted and averaged) . . . . **17·4**
Tube 2.—Phagocytic count (bacteria in 26 P.W.B.C. counted and averaged) . . . . **19·9**

B.

The patient's serum, 3 vols.: staphylococcus suspension, 1 vol.; the patient's shed corpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 15 P.W.B.C. counted and averaged) **35**
Tube 2.— Do. do. **36**

C.

The patient's serum, 3 vols.; staphylococcus suspension, 1 vol.; A. E. W.'s shed corpuscles, 3 vols.

Tube 1.—Phagocytic count (bacteria in 15 P.W.B.C. counted and averaged) **30**
Tube 2.— Do. do. **26**

Further Observations on the Rôle of the Blooc Fluids in connexion with Phagocytosis.[1]

By A. E. Wright and Stewart R. Douglas.

From the Laboratory of the Pathological Department, St. Mary's Hospita London, W.

Relation of the Opsonic Power of Human Blood to the Capacity of Resisting Invasio by the Staphylococcus Pyogenes—Experiments on the Opsonic Power of Huma Blood in its Relation to the Bacillus of Plague—Experiments on the Opsoni Power of Human Blood in Relation to Micrococcus Melitensis—Experiment on the Opsonic Power of Human Blood in Relation to the Bacillus Dysentericu (Shiga)—Experiments on the Opsonic Power of Human Blood in its Relatio to the Bacillus coli—Experiments on the Opsonic Power of Human Blood in it Relation to the Pneumococcus of Fraenkel—Experiments on the Opsonic Powe of Human Blood in its Relation to the Bacillus of Anthrax—Opsonic Power c Human Blood in its Relation to the Bacillus typhosus and the Cholera Vibrio– Opsonic Power of Human Blood in its Relation to the Diphtheria Bacillus and th Xerosis Bacillus.

In a previous communication we showed that the phagocytosis whic occurs when cultures of the staphylococcus pyogenes are added t human blood, is directly dependent upon the presence of certain sub stances in the blood which exert a specific effect upon the bacteria. W suggested that the bacteriotropic substances here in question migh appropriately be denoted by the term "opsonins."

In the present paper we propose to bring out certain further point in connexion with the "opsonic power" of the blood.

Relation of the Opsonic Power of Human Blood to the Capacity of Resist ing Invasion by the Staphylococcus Pyogenes.

It has already been shown[2] by one of us that patients who ar the subjects of acne, sycosis, or boils are characterized by a defectiv phagocytic power for the staphylococcus pyogenes. We have recentl been able to satisfy ourselves that this defective phagocytosis is deper dent upon a defect of opsonic power.

It has also been shown by one of us that the cure of these bacteri infections, which can in almost every instance be achieved by the inocul tion of appropriate quantities of sterilized staphylococcus cultures, i

[1] Reprinted from the *Proceedings of the Royal Society*, Vol. lxxiii, 1904.

[2] *Lancet*, March 29, 1902 (pp. 99 *et seq.*).

ociated with the acquirement of an increased phagocytic power. We ve now succeeded in establishing the fact—already adumbrated in r previous paper—that the increased phagocytosis which is associated th the achievement of the condition of immunisation here in question dependent, not upon a modification of the white corpuscles, but upon development of opsonins in the blood fluids.

The results of the subjoined experiment bring out this fact into clear ief.

Details of the Experiment.

Immunised Patient's Blood.—The patient, F.F., who had long been subject of aggravated staphylococcic sycosis, had, after prolonged d ineffectual treatment with antiseptics, been subjected to three ccessive inoculations of a sterilized staphylococcus culture. Under se inoculations his clinical condition had ameliorated itself in astonishing manner, and his phagocytic power, which had evious to the date of inoculation been less by half than that of a rmal man who served as a control, had increased in a progressive nner after each inoculation.

A sample of blood was now (by the technique elsewhere described) [1] awn off and mixed with $\frac{1}{10}$th of its volume of 10 per cent. citrate of da. A second sample of blood was drawn off and allowed to clot in ordinary way.

In the case of the first sample of blood the corpuscles were isolated m the plasma by repeated washing with physiological salt solution, d centrifugalization. The corpuscles thus isolated are referred to ow as "washed corpuscles."

In the case of the second sample of blood the serum was simply arated from the corpuscles in the ordinary way by centrifugalization.

Control Blood from a Normal Man.—The blood which served as a ntrol was obtained from a normal healthy man. It was drawn off in actly the same manner and was treated in each case by exactly the ne procedures as the blood obtained from the patient.

Bacterial Culture.—The bacterial culture employed in the experi- nts set forth below was obtained by suspending in physiological salt ution a portion of a twenty-four hours' growth of staphylococcus albus agar.

The quantities of serum, washed corpuscles, and staphylococcus ture which are specified below were then in each case taken up into apillary tube, mixed on a glass slide, re-aspirated into the tube, and ested together at blood heat for fifteen minutes. Films were then made d stained by Leishman's stain. Finally the number of ingested bacteria re enumerated in a series of polynuclear W.B.C. taken in order as y came.

[1] *Lancet*, January 23, 1904.

The phagocytic count given below—and the same applies throughou this paper—represents in each case the average number of bacteri ingested by the individual P.W.B.C. The number of polynuclear whit blood corpuscles which were counted is in each case inserted in brackets :—

Experiment.

A.

Immunised patient's washed corpuscles . . .	3 vols.
Immunised patient's serum	3 ,,
Suspension of staphylococcus culture . . .	1 vol.

Phagocytic count (20 P.W.B.C.). **25·7.**

B.

Washed corpuscles from normal man	3 vols.
Serum from normal man	3 ,,
Suspension of staphylococcus culture	1 vol.

Phagocytic count (15 P.W.B.C.). **13.**

C.

Immunised patient's washed corpuscles . . .	3 vols.
Serum from normal man	3 ,,
Suspension of staphylococcus culture . . .	1 vol.

Phagocytic count (15 P.W.B.C.). **13.**

D.

Washed corpuscles from normal man	3 vols.
Serum from immunised patient	3 ,,
Suspension of staphylococcus culture	1 vol.

Phagocytic count (15 P.W.B.C.). **28·2.**

Experiments on the Opsonic Power of Human Blood in its Relation t the Bacillus of Plague.

In these and all subsequent experiments, unless where otherwis specified, the technique employed was exactly the same as that employe in the experiments set forth above. It may further be premised tha the bacterial suspensions employed were in each case suspensions very young agar cultures—in most cases 24-hour cultures—in physi logical salt solution. By the term "heated serum" is in each case to b understood serum which has been subjected to a temperature of 60° for ten minutes or more.

Experiment 1.

A.

S. R. D.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of plague bacillus	1 vol.

Phagocytic count (20 P.W.B.C.), **3·0.**

B.

S. R. D.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of plague bacillus	1 vol.

Phagocytic count (25 P.W.B.C.), **0·7.**

Experiment 2.

A.

S. R. D.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of plague bacillus	1 vol.

Phagocytic count (20 P.W.B.C.), **13·1.**

B.

S. R. D.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of plague bacillus	1 vol.

Phagocytic count (20 P.W.B.C.), **2·1.**

Experiment 3.

A.

A. E. W.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of plague bacillus	1 vol.

Phagocytic count (21 P.W.B.C.), **19·6.**

B.

A. E. W.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of plague bacillus	1 vol.

Phagocytic count (54 P.W.B.C.), **8·4.**

Experiment 4.

A.

B. H. S.'s unheated serum	2 vols.
A. E. W.'s washed corpuscles	2 „
Suspension of plague bacillus	1 vol.

Phagocytic count (43 P.W.B.C.), **5·3.**

B.

B. H. S.'s heated serum	2 vols.
A. E. W.'s washed corpuscles	2 „
Suspension of plague bacillus	1 vol.

Phagocytic count (43 P.W.B.C.), **1·4.**

It may incidentally be noted in connexion with these experiments at while the plague bacilli which lay free in the films were in each case ite unaltered, many of these which had been ingested showed extremely aracteristic involution forms[1] such as we have not seen since we orked with freshly isolated plague cultures in Bombay in connexion th the Indian Plague Commission. So typical were the involution rms of the ingested plague bacilli, that we should not hesitate to employ e method of phagocytosis as an aid to diagnosis in the case of a doubtful ague culture.

[1] It may be observed that our plague culture—like other plague cultures which ve been cultivated on artificial nutrient media for a number of generations— s altogether lost the property of developing in a spontaneous manner the involu- n forms which are characteristic of freshly isolated plague cultures.

Experiments on the Opsonic Power of Human Blood in Relation to Micrococcus Melitensis.

Experiment 1.

A.

S. R. D.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of *Micrococcus Melitensis* . . .	1 vol.

Phagocytic count (10 P.W.B.C.), **26·9.**

B.

S. R. D.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of *Micrococcus Melitensis* . . .	1 vol.

Phagocytic count (10 P.W.B.C.), **9·2.**

Experiment 2.

A.

A. E. W.'s unheated serum	3 vols.
A. E. W.'s washed corpuscles	3 ,,
Suspension of *Micrococcus Melitensis* . . .	1 vol.

Phagocytic count (21 P.W.B.C.), **10·0.**

B.

A. E. W.'s heated serum	3 vols.
A. E. W.'s washed corpuscles	3 ,,
Suspension of *Micrococcus Melitensis* . . .	1 vol.

Phagocytic count (21 P.W.B.C.), **2·4.**

Experiment 3.

A.

S. R. D.'s heated serum	3 vols.
A. E. W.'s washed corpuscles	3 ,,
Suspension of *Micrococcus Melitensis* . . .	1 vol.

Phagocytic count (21 P.W.B.C.), **12·9.**

B.

S. R. D.'s heated serum	3 vols
A. E. W.'s washed corpuscles	3 ,,
Suspension of *Micrococcus Melitensis.* . . .	1 vol.

Phagocytic count (21 P.W.B.C.), **0·9.**

Experiments on the Opsonic Power of Human Blood in Relation to the Bacillus Dysentericus (Shiga).

Experiment 1.

A.

S. R. D.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of Shiga's bacillus	1 vol.

Phagocytic count (20 W.P.B.C.), **4·2.**

B.

S. R. D.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of Shiga's bacillus	1 vol.

Phagocytic count (20 P.W.B.C.), **0·0.**

Experiment 2.

A.

A. E. W.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of Shiga's bacillus	1 vol.

Phagocytic count (20 P.W.B.C.), **5·4.**

B.

A. E. W.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of Shiga's bacillus	1 vol.

Phagocytic count (33 P.W.B.C.), **0·1.**

Experiment 3.

A.

S. R. D.'s unheated serum	2 vols.
S. R. D.'s washed corpuscles	2 ,,
Suspension of Shiga's bacillus	1 vol.

Phagocytic count (20 P.W.B.C.), **3·6.**

B.

S. R. D.'s heated serum	2 vols.
S. R. D.'s washed corpuscles	2 ,,
Suspension of Shiga's bacillus	1 vol.

Phagocytic count (20 P.W.B.C.), **0·2.**

A certain number of the bacilli (and these bacilli were found indiffer-
ly in the interior of the cells and free in the preparation) had, in the
e of the experiments undertaken with unheated serum, undergone
ierulation.

periments on the Opsonic Power of Human Blood in its Relation to the Bacillus coli.

Experiment 1.

A.

B. H. S.'s unheated serum	3 vols.
B. H. S.'s washed corpuscles	3 ,,
Suspension of the *Bacillus coli*	1 vol.

Phagocytic count (20 P.W.B.C.), **3·8.**

B.

B. H. S.'s heated serum	3 vols.
B. H. S.'s washed corpuscles	3 ,,
Suspension of the *Bacillus coli*	1 vol.

Phagocytic count (20 P.W.B.C.), **0·75.**

Experiment 2.

A.

F. F.'s unheated serum	3 vols.
F. F.'s washed corpuscles	3 ,,
Suspension of the *Bacillus coli*	1 vol.

Phagocytic count (20 P.W.B.C.), **5.**

B.

F. F.'s heated serum	3 vols.
F. F.'s washed corpuscles	3 ,,
Suspension of the *Bacillus coli*	1 vol.

Phagocytic count (21 P.W.B.C.), **0·76.**

Experiments on the Opsonic Power of Human Blood in its Relation to the Pneumococcus of Fraenkel.

Experiment 1.

A.

S. R. D.'s unheated serum	2 vols.
S. R. D.'s washed corpuscles	2 „
Suspension of the pneumococcus of Fraenkel . .	1 vol.

Phagocytic count (15 P.W.B.C.), **16.**

B.

S. R. D.'s heated serum	2 vols.
S. R. D.'s washed corpuscles	2 „
Suspension of Fraenkel's pneumococcus . . .	1 vol.

Phagocytic count (40 P.W.B.C.), **1·1.**

Experiment 2.

A.

A. E. W.'s unheated serum	2 vols.
S. R. D.'s washed corpuscles	2 „
Suspension of Fraenkel's pneumococcus . . .	1 vol.

Phagocytic count (23 P.W.B.C.), **6.**

B.

A. E. W.'s heated serum	2 vols.
S. R. D.'s washed corpuscles	2 „
Suspension of Fraenkel's pneumococcus . . .	1 vol.

Phagocytic count (40 P.W.B.C.), **0·2.**

Experiments on the Opsonic Power of Human Blood in its Relation to the Bacillus of Anthrax.

Experiment 1.

A.

S. R. D.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of *Bacillus anthracis*	1 vol.

Enumeration was here impossible, but there was everywhere evidence of phagocytosis. In the few cases where the leucocytes had not ingested bacteria, they were found to have extended themselves in a characteristic grasping manner along the bacterial threads (Plate 5).

B.

S. R. D.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of the *Bacillus anthracis*	1 vol.

Here there were practically no signs of phagocytosis. The cells were everywhere empty, and they had not drawn themselves into intimate contact with the anthrax threads (Plate 6).

Experiment 2.

A.

A. E. W.'s unheated serum	2 vols.
S. R. D.'s washed corpuscles	2 „
Broth culture of anthrax	1 vol.

Phagocytic count (36 P.W.B.C.), 2·4 (approximate only).

B.

A. E. W.'s heated serum	2 vols.
S. R. D.'s washed corpuscles	2 „
Broth culture of anthrax	1 vol.

Phagocytic count (100 P.W.B.C.), **0.**

onic Power of Human Blood in its Relation to the Bacillus typhosus and the Cholera Vibrio.

It is well known that human blood exerts a very considerable bacteri-
l power upon cultures of the *Bacillus typhosus* and of the cholera
io. The destructive effect in question manifests itself to microscopical
rvation in the form of very profound morphological changes which
e under observation in cultures which have been digested with
eated serum. The bacteria in such cultures, after undergoing agglutin-
n and spherulation, swell up and lose their chemical affinity for aniline
s. Finally they are completely dissolved.

t is manifest that where disintegrative changes of this kind are occur-
under the influence of the serum, opsonic effects will be more or less
st into the background. These last will, in the case of phagocytic
riments conducted with unheated serum, be masked, on the one
d, by the fact that there will be fewer bacteria available for phagocy-
, and on the other hand by the fact that intracellular disintegration
it may be presumed, be more rapid in the case where the serum has
dy exerted a disintegrating effect on the bacteria anterior to their
stion.

astly, ingested bacteria which have lost their characteristic chemical
ity for their stain may readily escape enumeration.

ll these points must be taken into consideration in connexion with
subjoined experiments :—

Experiment 1.

A.

S. R. D.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of the cholera vibrio	2 „

Everywhere considerable phagocytosis. Complete spherulation of
st all the micro-organisms within and all the micro-organisms out-
the cells. No indication of vacuolation round the ingested bacteria
te 3).

Phagocytic count (14 P.W.B.C.), **24** (*circ.*).

B.

S. R. D.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of the cholera vibrio	2 „

Everywhere considerable phagocytosis. No spherulation of the

micro-organisms either within or without the leucocytes. Very mark vacuolation of the leucocytes round the ingested bacteria (fig. 4)

Phagocytic count (11 P.W.B.C.), **26·2** (*circ.*).

Experiment 2.

A.

A. E. W.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of the cholera vibrio	1 vol.

Complete spherulation of all the bacteria, whether within or with the cells.

Phagocytic count (21 P.W.B.C.), **8·1** (*circ.*).

B.

A. E. W.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Suspension of the cholera vibrio	1 vol.

No spherulation of the micro-organisms, either within or without leucocytes.

Phagocytic count (13 P.W.B.C.), **0·8.**

Experiment 3.

A.

S. R. D.'s unheated serum	2 vols.
S. R. D.'s washed corpuscles	2 ,,
Broth culture of the typhoid bacillus	2 ,,

Much phagocytosis. Complete spherulation of all the extracellu micro-organisms. Many of the bacilli in the interior of the leucocy have completely preserved their original contours, others—probal the later ingested ones—are spherulated (Plate 1).

B.

S. R. D.'s heated serum	2 vols.
S. R. D.'s washed corpuscles	2 ,,
Broth culture of the typhoid bacillus	2 ,,

Much phagocytosis. All the micro-organisms, whether within without the leucocytes, are morphologically unaltered and have preserv their staining properties unimpaired (Plate 2).

Experiment 4.

A.

A. E. W.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 ,,
Broth culture of the typhoid bacillus	2 ,,

Complete spherulation of all the extracellular bacteria which ha escaped solution. In interior of leucocytes most of the bacteria ha undergone spherulation, but in the centre of the corpuscles som

obably those which were soonest ingested—are morphologically un-
ered and preserve their staining properties unaltered.

Phagocytic count, 100 (estimated).

B.

A. E. W.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Broth cultivation of the typhoid bacillus . . .	1 vol.

No spherulation, either within or without the cells.

Phagocytic count (20 P.W.B.C.), 31·8 (*circ.*).

Experiment 5.

A.

S. R. D.'s unheated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of the typhoid bacillus	1 vol.

All the bacilli both within and without the cells have undergone
ierulation.

Phagocytic count (11 P.W.B.C.), 13·6.

B.

S. R. D.'s heated serum	3 vols.
S. R. D.'s washed corpuscles	3 „
Suspension of the typhoid bacillus	1 vol.

No spherulation either within or without the leucocytes.

Phagocytic count (23 P.W.B.C.), 7·2.

Of incidental interest in connexion with the above experiments is
demonstration which they afford, that the spherulation of the intra-
ular ingested micro-organisms, which has been often ascribed to the
ncy of the leucocytes, is in reality due to agency of the blood fluids.

sonic Power of Human Blood in its Relation to the Diphtheria Bacillus and the Xerosis Bacillus.

Experiment 1.

A.

A. E. W.'s unheated serum	3 vols.
A. E. W.'s washed corpuscles	3 „
Suspension of the diphtheria bacillus	3 „

Phagocytic count (27 P.W.B.C.), 0·7.

B.

A. E. W.'s heated serum	3 vols.
A. E. W's washed corpuscles	3 „
Suspension of the diphtheria bacillus	3 „

Phagocytic count (29 P.W.B.C.), 4·1.

Experiment 2.

A.

B. H. S.'s unheated serum	3 vols.
B. H. S.'s washed corpuscles	3 „
Suspension of the diphtheria bacillus	2 „

Phagocytic count (20 P.W.B.C.), **8·0.**

B.

B. H. S.'s heated serum	3 vols.
B. H. S.'s washed corpuscles	3 „
Suspension of the diphtheria bacillus	2 „

Phagocytic count (20 P.W.B.C.), **10·9.**

Experiment 3.

A.

B. H. S.'s unheated serum	3 vols.
B. H. S.'s washed corpuscles	3 „
Suspension of the diphtheria bacillus	1 vol.

Phagocytic count (44 P.W.B.C.), **4·0.**

B.

B. H. S.'s heated serum	3 vols.
B. H. S.'s washed corpuscles	3 „
Suspension of the diphtheria bacillus	1 vol.

Phagocytic count (50 P.W.B.C.), **8·3.**

Experiment 4.

A.

A. E. W.'s unheated serum	3 vols.
E. A. W.'s washed corpuscles	3 „
Suspension of the xerosis bacillus	1 vol.

Phagocytic count (40 P.W.B.C.), **2·8.**

B.

A. E. W.'s heated serum	3 vols.
A. E. W.'s washed corpuscles	3 „
Suspension of the xerosis bacillus	1 vol.

Phagocytic count (25 P.W.B.C.), **3·2.**

Experiment 5.

A.

B. H. S.'s unheated serum	3 vols.
B. H. S.'s washed corpuscles	3 „
Suspension of the xerosis bacillus	1 vol.

Phagocytic count (30 P.W.B.C.), **6·3.**

B.

B. H. S.'s heated serum	3 vols.
B. H. S.'s washed corpuscles	3 „
Suspension of the xerosis bacillus	1 vol.

Phagocytic count (30 P.W.B.C.), **6.**

Conclusions.

The experimental data which have been set forth above esta
that the opsonic action of the blood fluids—to which attention wa
the first time directed in our previous communication—is exerted

usively upon the *Staphylococcus pyogenes*, but also upon the *Bacillus* s, the *Micrococcus Melitensis*, the *Diplococcus pneumoniae* of Fraenkel, *Bacillus coli*, the *Bacillus dysenteriae* (Shiga), the *Bacillus anthracis*, *Bacillus typhosus*, and the *Vibrio cholerae Asiaticae*.

o far as we have gone, the *Bacillus diphtheriae* and its congener the *llus xerosis* have proved to be the only pathogenetic bacteria which insensible to this action of the blood fluids.

'aking these experimental data in conjunction with other facts h have been elicited by us, or as the case may be by one of us working connexion with Captain F. Windsor,[1] I.M.S., with regard to the ericidal action exerted by human blood upon the various species athogenetic micro-organisms, we may classify these bacteria in the wing categories :—

1) *Bacteria which are eminently sensible to the bactericidal, bacteriolytic, opsonic action of normal human blood fluids.*—The *Bacillus typhosus* the *Vibrio cholerae Asiaticae.*

2) *Bacteria which are in some measure sensible to the bactericidal action e normal human blood fluids, and which are eminently sensible to its nic action.*—The *Bacillus coli* and the *Bacillus dysenteriae.*

3) *Bacteria which are absolutely insensible to the bactericidal action he normal human blood fluids, but are eminently sensible to the nic action of these fluids.*—The *Staphylococcus pyogenes*, the *Bacillus* s, the *Micrococcus Melitensis*, the *Diplococcus pneumoniae* of Fraenkel.

4) *Bacteria which are insensible both to the bactericidal and to the nic action of the normal human blood fluids.*—The *Bacillus diphtheriae Bacillus xerosis.*

t may be pointed out in conclusion that the demonstration furnished e, that successful immunisation against the staphylococcus pyogenes pendent upon an elaboration of opsonins in the system of the inocu- l patient, suggests that successful immunisation against plague and a fever, and we may add against streptococcal invasions, may be vise dependent upon the elaboration of opsonins.

t will be manifest that if this is so, the determination of the opsonic er of the blood is calculated to render services also in connexion with testing of any therapeutic sera which may find an application in exion with the disease.

[1] *Journal of Hygiene, loc. cit.* (*vide supra*, pp. 45–72).

On the Action exerted upon the *Staphylococ* *pyogenes* by Human Blood Fluids, and the Elaboration of Protective Elements the Human Organism in Response to oculations of a Staphylococcus Vaccine

By A. E. Wright and Stewart R. Douglas.

From the Laboratory of the Pathological Department, St. Mary's Hos London, W.

1. Nature of the Action which is exerted upon the Staphylococcus pyogenes by No Blood Fluids, and by the Blood Fluids of Patients inoculated with a Staphyloc Vaccine—2. Comparison of the Phagocytic Power of the Subjects of Sta lococcus Invasion with the Phagocytic Power of Normal Persons—3. Or Distribution of Opsonins in the Infected Organism—4. Determination o Question as to whether the Opsonins which come into Consideration in Conn with the Protection of the Organism against Staphylococcus Invasion are pr in the Blood of the Infant at Birth—5. Determination of the Course o Reaction of Immunisation obtained in response to Inoculations of a Staphyloc Vaccine.

The subject matter with which we have here to deal may be distrib under the following headings :—

(1) Determination of the nature of the action which is exerted the *Staphylococcus pyogenes* by normal human blood fluids, and the blood fluids of patients who have been inoculated with a sta lococcus vaccine.

(2) Comparison of the phagocytic power of the subjects of sta lococcus invasion with the phagocytic power of normal individua

(3) Distribution in the infected organism of the opsonins which come into consideration.

(4) Determination of the question as to whether the opso are present in the blood of the infant at birth.

(5) Determination of the course of the reaction of immunis which supervenes upon the inoculation of a staphylococcus vacci

1. Nature of the Action which is exerted upon the Staphylococcus pyog by Normal Blood Fluids, and by the Blood Fluids of Patients in lated with a Staphylococcus Vaccine.

Bactericidal Action.—It was shown in the course of the cla

[1] Reprinted from the *Proceedings of the Royal Society*, vol. lxxiv, 1904.

rches on the bactericidal power of the blood which were conducted Tuttall [1] in Flügge's laboratory, that the staphylococcus offers resist- to the bactericidal action of the blood fluids. Two years afterwards observations of Nuttall, which had reference to the blood of animals, extended by Stern [2] to human blood. The methods employed by all and Stern alike did not, however, permit of a comparatively l bactericidal action being distinguished from a complete absence of ericidal action.

ie question as to how far the staphylococcus offers resistance to bactericidal action of human blood was reinvestigated by one of he results being published (*a*) in a paper dealing with anti-staphy- cus inoculations,[3] and (*b*) in a paper written in conjunction with ain F. Windsor, I.M.S.,[4] on the bactericidal action exerted by human d upon a variety of pathogenic micro-organisms. It was established ie researches here in question, which were conducted with the more ate methods of investigation set forth in the *Proceedings of the l Society* [5] and in the *Lancet* [6] respectively, (*a*) that normal an blood does not exert upon the staphylococcus any bacteri- action whatever, and (*b*) that anti-staphylococcus inoculations not lead to a development of any bactericidal power in the d.[7]

psonic Action.—It having become evident in the course of these rches that the effect of anti-staphylococcus inoculation is not e found in a development of bactericidal properties in the blood s, attention was directed to the measurement of the phagocytic r of the blood. Taking to aid the method of phagocytic estimation sed by Major W. B. Leishman, R.A.M.C., who was then our fellow- ker, it was ascertained that successful immunisation against staphy- cus goes in every case hand in hand with the acquirement of in- sed phagocytic power.

ertain difficulties having suggested themselves in connexion with attribution of this result to a "training" of the white blood corpuscles, addressed ourselves to a further investigation of the phenomena hagocytosis.

n the course of this investigation [8] it became clear that phagocytosis acteria is dependent upon an effect exerted upon the bacteria by blood fluids. We spoke of this effect as an "opsonic effect."

n a second research,[9] in which we extended our previous observations,

Nuttall, *Zeitschrift f. Hygiene*, 1888, vol. iv.
Stern, *Verhandlungen des IX Congresses f. Innere Medicin*, 1890.
Wright, *Lancet*, March 29, 1902 (pp. 99 *et seq.*) .
Wright and Windsor, *Journal of Hygiene*, vol. ii, No. 4, March, 1902 (pp. 45– *pra*).
Wright, *Roy. Soc. Proc.*, vol. 71, 1902.
Wright, *Lancet*, December 1, 1900, and March 2, 1901.
Vide Journal of Hygiene (loc. cit.), Tables VII and VIII. (pp. 56–57 *supra*).
Wright and Douglas, *Roy. Soc. Proc.*, 1903, vol. 72 (pp. 75–87 *supra*).
Wright and Douglas, *Roy. Soc. Proc.*, 1904, vol. 73 (pp. 88–89 *supra*).

on the opsonic power of the blood fluids, we showed that the increa phagocytic effect which is obtained with the blood of successfully imm ised persons is attributable not to any modification induced in the l cocytes, but to an increased opsonic power in the blood fluids. Conclus evidence of this was obtained by separating, in the case of two blo of conspicuously different phagocytic power in each case, the blood flu from the corpuscular elements, and then effecting an interchange of blood fluids. The leucocytes of the successfully immunised pati exhibited under these circumstances the smaller phagocytic act characteristic of the blood of the normal individual who served a control, while the leucocytes of the normal individual exhibited increased phagocytic action characteristic of the blood of the successfu immunised patient. (See pp. 89–90.)

The witness of the experiment just referred to, and of a previ experiment incorporated in our first paper, is confirmed by similar resu obtained in connexion with the tubercle bacillus. (See p. 117.)

Agglutinating Action.—Normal human serum does not exert any ch acteristic agglutinating action upon the staphylococcus. Such agg tination as is obtained is not very sensibly increased under the influe of staphylococcus inoculations.

2. Comparison of the Phagocytic Power of the Subjects of Staphylococ Invasion with the Phagocytic Power of Normal Persons.

It is clear from what has been said above that the essential cha which takes place in human blood, as a result of the inoculation of stap lococcus cultures, is an increase in the phagocytic power, depend upon an increase of the opsonic elements in the blood.

Further evidence of the essential importance of the phagocytic opsonic power in connexion with resistance to staphylococcus invasi is obtained by contrasting the phagocytic power of the subjects of stap lococcus invasion with that of normal individuals.

Our observations on this subject were made in some instances comparing the phagocytic power of the decalcified blood of the pati with the phagocytic power of the decalcified blood of a normal pers More frequently we employed in our experiments, respectively, patient's serum and the serum of a normal person in each case in asso tion with the washed corpuscles derived from a normal man.

The results of our observations are tabulated below :—

·le showing the Ratio in which the Phagocytic or Opsonic Power of the Patients Blood stood in each case to the Phagocytic or Opsonic Power of the Normal Individual who furnished the Control Blood.

(The phagocytic power of the control blood is taken in each case as unity.)

itials of Patient.	Form of Staphylococcus Invasion.	Phagocytic or Opsonic Index.
G.	Furunculosis	0·48
F.	Sycosis	0·49
E.	Acne	0·64
H.	Furunculosis	0·87
B.	Acne	0·55
H.	"	0·82
H.	Furunculosis	0·79
G.	"	0·7
L.	Acne and sycosis	0·74
C.	Furunculosis	0·87
L.	"	0·88
P.	"	0·39
F.	Very aggravated sycosis	0·1
F. D.	Acne	0·73
C.	Sycosis	0·8
M.	Acne	0·48
M.	Sycosis	0·37
P.	Acne	0·6
S.	Pustular affection of lips	0·6
V.	Repeated septic infection	0·67

In view of these observations and of the fact that we have not come ·oss any instance of the association of a normal phagocytic power th a staphylococcus infection,[1] the conclusion would seem justified at a low phagocytic power and staphylococcus infection are related each other by some fact of causation. While it is *à priori* possible at the diminished phagocytic power which characterizes those infected the staphylococcus might be the result of the staphylococcus invasion, is infinitely more probable, in view of the entire absence of clinical nptoms in the slighter cases of staphylococcus infection, that it is e defective phagocytic power of the patient which furnishes to the phylococcus which is normally present upon the surface of the body e opportunity for invading the skin.

It is shown elsewhere (see p. 118) that a similar problem arises in nnexion with the circumstance that a low phagocytic power, with pect to the tubercle bacillus, is generally found in association with bercular infection.[2]

3. On the Distribution of Opsonins in the Infected Organisms.

It is a fundamentally important but unappreciated fact in connexion th bacterial infections that the *bacteriotropic pressure*—we designate

[1] *We have since come across such instances. They are, however, exceptional.*
[2] *We had here in view strictly localized tubercular infections.*

by this term the mass effect exerted upon the invading bacteria by th anti-bacterial substances contained in the blood fluids—does not stan at the same level in every part of the infected organism.

One of us has, in conjunction with Captain George Lamb, I.M.S. demonstrated in the case of patients who had succumbed respective to typhoid and Malta fever that the amount of agglutinins in the splen pulp is invariably less,[2] in some instances over 200 times less, than the circulating blood. It was further shown in the paper in questio that there was a similar difference as between the fluid obtained fro the typhoid spots and the fluid of the circulating blood. Captain Lamb gave a further extension to these observations by demonstrating, the case of monkeys, examined immediately after the crisis spirillum fever, that the splenic pulp (where the spirilla still survi after they have disappeared from the circulation) is much poorer bactericidal and bacteriolytic substances than the circulating blood.

It is shown by these observations that the *Bacillus typhosus*, th *Micrococcus Melitensis* and the *Spirillum Obermeyeri*, respectively multipl or, as the case may be, maintain their existence, within the infecte organism in regions of low bacteriotropic pressure. We may legitimate assume that the lowered bacteriotropic pressure in the nidus, whe the micro-organisms are cultivating themselves, results from a retarde replacement of anti-bacterial substances which are removed from the bod fluids where these come into contact with bacteria.

Influenced by the results of the observations which have been ju set out, we have addressed ourselves to the task of investigating th distribution of the opsonins in the case where the human organis is invaded by the staphylococcus. With this intent we have institute comparisons between the serum obtained from the circulating bloo and the fluid obtained by centrifugalization, from pus. It will be see from the observations set forth below that what has been shown to ho true with respect to the distribution of agglutinins and bactericidal an bacteriolytic substances respectively in the bacterial infections befor mentioned, holds true also in the case of the opsonins in the case staphylococcic infection. In view of this fact, and of the similar fac which we set out elsewhere in connexion with tubercular infection (se pp. 119–120), it may be enunciated as a proposition of general applic tion that the invading micro-organisms cultivate themselves in th organism in regions of lowered bacteriotropic pressure.

Case 1.

13.4.04. Patient with an alveolar abscess pointing on the chee Pus gives a pure culture of staphylococcus.

[1] Wright and Lamb, *Lancet*, December 23, 1898 (pp. 36–44 *supra*).

[2] This observation, so far as it applies to typhoid, had been anticipated by Pa Courmont, *Soc. de Biologie*, February 20 and March 28, 1897.

[3] Lamb, *Scientific Memoirs by Officers of the Medical and Sanitary Departmen of the Government of India*, vol. xii, pp. 96 *et seq.*

A.

Patient's serum	2 vols.
A. E. W.'s washed corpuscles	2 „
Staphylococcus emulsion	1 vol.

Phagocytic count [1] (average of 20 P.W.B.C.), **30·3.**

B.

Supernatant fluid from pus	2 vols.
A. E. W.'s washed corpuscles	2 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 20 P.W.B.C.), **5·1.**

tio of phagocytic count of serum to phagocytic count of supernatant fluid from 1 : 0·17.

15.4.04. Patient has had fomentations applied to cheek since abscess opened on 13.4.04. Abscess rapidly healing.

A.

Patient's serum	2 vols.
A. E. W.'s washed corpuscles	2 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 20 P.W.B.C.), **10·05.**

B.

Supernatant fluid from pus	2 vols.
A. E. W.'s washed corpuscles	2 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 20 P.W.B.C.), **10·1.**

Ratio of phagocytic count of serum to phagocytic count of supernatant fluid of 1 : 1.

Case 2.

Patient with patellar abscess. Pus from abscess furnishes a pure vth of streptococcus.

A.

Patient's serum	2 vols.
A. E. W.'s washed corpuscles	2 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 20 P.W.B.C.), **14·2.**

B.

Supernatant fluid of pus	2 vols.
A. E. W.'s washed corpuscles	2 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 40 P.W.B.C.), **1·25.**

tio of phagocytic count of serum to phagocytic count of supernatant fluid of pus, ·09.

Determination of the Question as to whether the Opsonins which come into Consideration in Connexion with the Protection of the Organism against Staphylococcus Invasion are present in the Blood of the Infant at Birth.

Opportunity offering, we have thought it worth while to determine ther the protective substances which come into consideration in

[1] The phagocytic count was here, as elsewhere, determined by counting the ber of bacteria ingested in the specified number of polynuclear leucocytes after sting together in a capillary tube for 15 mins. at 37° C. the serum, corpuscles, bacterial suspension.

connexion with the Staphylococcus pyogenes are present in the bl
at birth. For this purpose we have made a series of comparative esti
tions of the opsonic power of the blood of child and mother, employ
for this purpose respectively placental blood and blood drawn off dire
from the mother immediately after the completion of parturition.
are indebted to Messrs. B. H. Spilsbury and J. Freeman for the collect
of the bloods. The observations we have made are as follows :—

Observations.

Blood drawn off, in the case of the mother from the finger ; in the c
of the child, from the umbilical cord.

No. 1.

A.

Mother's serum	3 vols.
A. E. W.'s washed corpuscles	3 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 20 P.W.B.C.), **15·1.**

B.

Infant's serum	3 vols.
A. E. W.'s washed corpuscles	3 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 20 P.W.B.C.), **16·5.**

No. 2.

A.

Mother's serum	2 vols.
A. E. W.'s washed corpuscles	2 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 20 P.W.B.C.), **12·65.**

B.

Infant's serum	2 vols.
A. E. W.'s washed corpuscles	2 „
Staphylococcus emulsion	1 vol.

Phagocytic count (average of 20 P.W.B.C.), **12·25.**

5. Determination of the Course of the Reaction of Immunisation obtai in response to Inoculations of a Staphylococcus Vaccine.

We have in a very considerable number of cases plotted out by
aid of the phagocytic method the course of the reaction of immunisat
which occurs in response to inoculations of a staphylococcus vaccine.

A preliminary word or two may be devoted to the description
the mode of preparation of the vaccine.

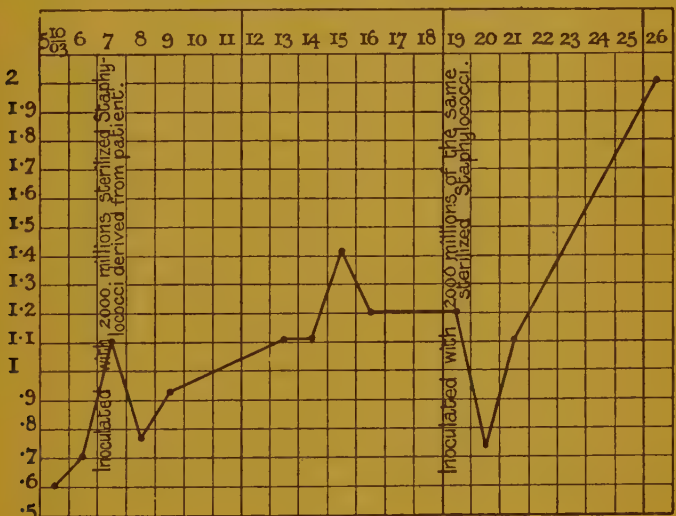
The procedure we adopt is as follows :—

We add to a 24-hours' growth of staphylococcus on sloped agar t
about 10 c.c. of sterile physiological salt solution. Churning up
culture with this, and letting it stand in order to allow all the unresol
bacterial masses to subside, we draw off the supernatant fluid by syp
action into a special form of tube, and heat to 60° C. for half an ho

We now place the tube in an incubator and incubate for 24 hours order to allow of a multiplication of any bacteria which may have vived the heating. We now take a sample of the vaccine and inoculate upon agar with a view to testing its sterility. Before heating, a sample the suspension has been drawn off for enumeration under the microscope the procedure described by one of us in the *Lancet* of July 5, 1902.

After verifying the sterility of the vaccine we now dilute with a suffincy of physiological salt solution to bring down the number of staphyocci in the cubic centimetre to 2,500,000,000. Finally we add lysol sufficient quantity to bring the content of the vaccine in this antiseptic 0·25 per cent.

In connexion with boils and sycosis a vaccine made from the *Staphyoccus aureus*; in cases of acne a vaccine made from a mixture of *iphylococcus albus* and *citreus* is appropriate.



Curve 1.

A dose of 0·5 to 1 c.c. of the vaccine made as above is a suitable antum for a first inoculation. For subsequent inoculations 1 to 2 c.c. the vaccine may be employed.[1]

Below are subjoined four of the more instructive of the curves which have obtained by the periodical examination of the phagocytic power the blood subsequent to inoculations of staphylococcus vaccines.

Curve 1.—The curve here in question applies to a medical man who d suffered from boils almost continuously for 4 years.

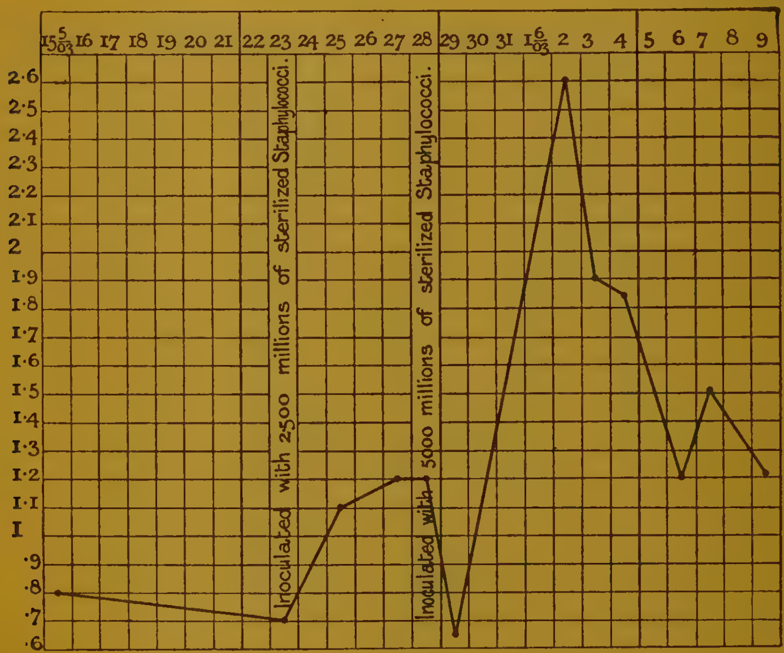
On the date when he presented himself for treatment he had two ils on his neck. It will be seen that the phagocytic index recorded for at day was 0·6—the phagocytic power of the normal control blood ing throughout taken as unity.

[1] *We have since found that much smaller doses may with advantage be employed.*

On the next day and the day subsequent phagocytic indices of 0
and 1·1 respectively were recorded. This altogether spontaneous improv
ment of the phagocytic power went hand in hand with a striking improv
ment in the condition of the boils.

The patient was now inoculated with a quantum of sterilized cultu
of staphylococcus corresponding to 2,000,000,000 of staphylococ
The culture employed was derived from the patient's boils.

On the day subsequent to inoculation the patient's phagocytic pow
was found to be reduced. Contemporaneously with the developme
of this "negative phase," an irritable pimple developed on the nec



Curve 2.

We may see in this, for it is a phenomenon which has manifested itse
again and again in this connexion with our inoculations, an indicatio
that the negative phase is associated with a diminished resisting pow
to invasion by the staphylococcus.

On the second day after the inoculation an improvement in th
phagocytic power was recorded. The "positive phase," which is he
heralded, reached its acme on the eighth day subsequent to inocul
tion.

On the twelfth day the patient was re-inoculated with the same qua
tum of vaccine as was employed on the first occasion. As on the previo
occasion, inoculation was followed by a negative, succeeded by a positi

hase. For a period of weeks after the inoculation, when the patient assed out of observation, he remained perfectly free from boils.

Curve 2.—This curve has reference to a patient who suffered from ggravated sycosis. A pure cultivation of *Staphylococcus citreus* was btained from the inflamed hair follicles. He had been treated without ppreciable benefit for seventeen months by antiseptics.

Reference to the curve will show that the patient's phagocytic power ith respect to the staphylococcus was here, as in the last case, less than hat of the normal man who served as a control.

After his phagocytic power had been twice observed, he was inoculated ith a quantum of sterilized staphylococcus culture corresponding to ,500,000,000 of staphylococci. These staphylococci were derived from he culture above referred to.

Subsequent to inoculation we have here upon the curve instead of a se preceded by a fall, only a rise. The absence of recorded negative hase is in all probability to be referred to the circumstance that two days ere intervened between the inoculation and the first subsequent blood xamination.

On the sixth day subsequent to inoculation the patient was reinculated with a double quantum of staphylococcus vaccine. This inoculaon was followed in a typical manner by a negative and positive phase. Vith respect to this last it will be seen that the curve attained its acme n the fifth day, and then declined in the usual manner.

Within a week after the second inoculation practically every trace f sycosis had disappeared. The patient was now lost sight of.

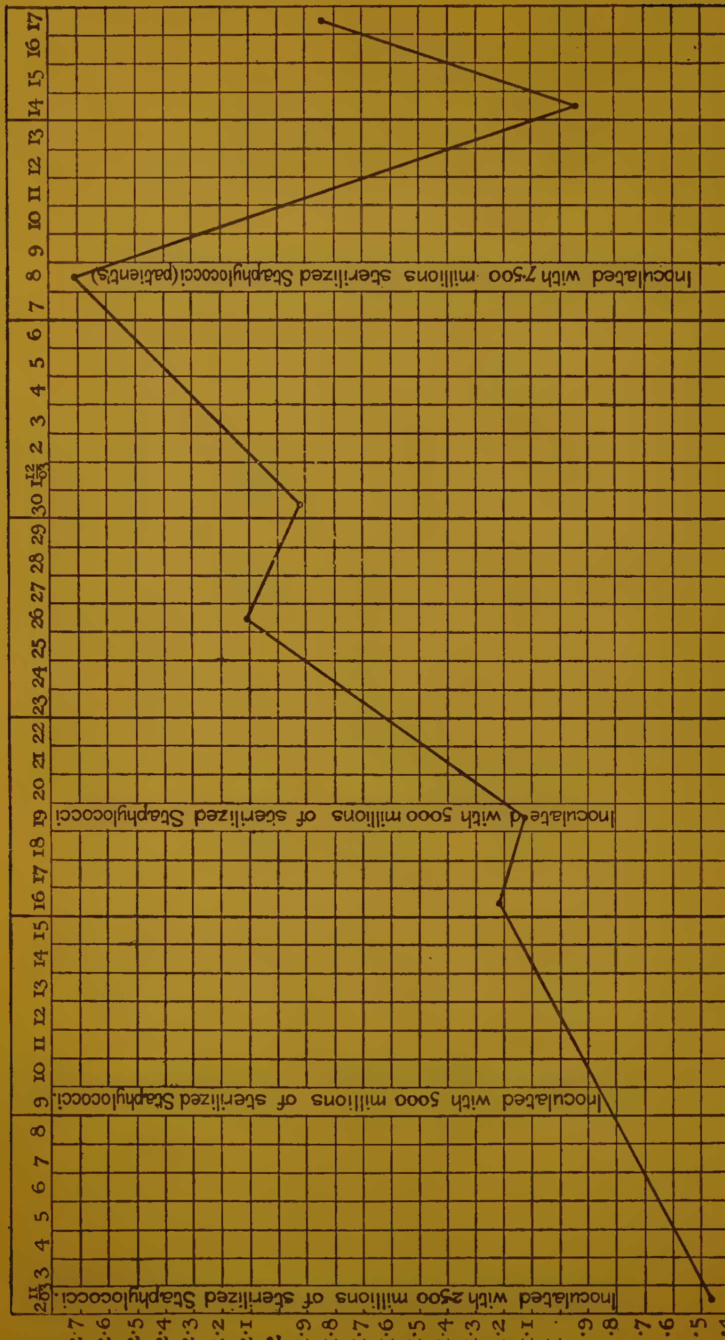
Curve 3.—The curve here in question applies to a labourer who was he subject of aggravated sycosis. He had suffered at intervals from hildhood from boils and other chronic staphylococcus infections. A ure cultivation of *Staphylococcus aureus* was obtained from the inflamed air follicles. He had been treated ineffectually for months by the sual methods.

As will be seen on reference to the chart, the phagocytic power of he blood was here investigated only from week to week instead of at ore frequent intervals. As a result the positive phase of the reaction s alone on record in the case of the first three inoculations. In the ase of the fourth inoculation—conducted with a larger quantum of he vaccine—the negative phase was still in evidence six days after the oculation.

The patient, who was all but completely cured at the date upon which he curve concludes, afterwards relapsed after free indulgence in alcohol. Ie is now, as a result of further inoculations, again practically well.

Curve 4.—This curve applies to a healthy man of 24 who, while in raining for a boat race, developed a boil on his gluteal region and subequently a crop of boils on his neck.

Reference to the curve will show that his phagocytic index stood t the date of his first inoculation at 0·84.

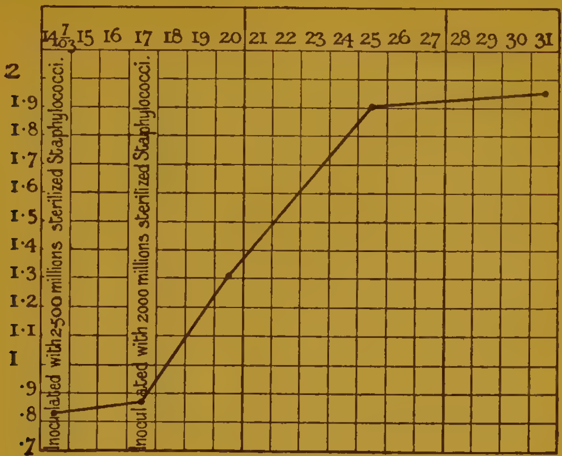

Inoculated with 2500 millions of sterilized Staphylococci.
Inoculated with 5000 millions of sterilized Staphylococci.
Inoculated with 5000 millions of sterilized Staphylococci
Inoculated with 7500 millions sterilized Staphylococci (patients)
2·7
2·6
2·5
2·4
2·3
2·2
2·1
2
1·9
1·8
1·7
1·6
1·5
1·4
1·3
1·2
1·1
1
·9
·8
·7
·6
·5
·4

Curve 3.

A quantum of sterilized staphylococcus culture corresponding to)0,000,000 of staphylococci was inoculated.

Three days afterwards his phagocytic index stood at 0·88.

A further quantum of 2,000,000,000 staphylococci was inoculated:



CURVE 4.

On the fifth and again on the eleventh day after inoculation the ient's phagocytic index stood respectively at 1·9 and 1·95.

Improvement in the patient's boils was already apparent at the date the second inoculation. After this they completely aborted.

The patient afterwards relapsed, but did not come up for further ervation.

On the Action exerted upon the Tuberc[...] Bacillus by Human Blood Fluids, and [...] the Elaboration of Protective Elements [...] the Human Organism in Response to In[...]culations of a Tubercle Vaccine.[1]

By A. E. Wright and Stewart R. Douglas.

From the Laboratory of the Pathological Department, St. Mary's Hosp[...] London, W.

1. On the Tuberculotropic Elements of Human Blood, and on the Content of Normal Blood in these Elements—Preliminary Experiments—2. Action exe[...] upon the Tubercle Bacillus by the Blood fluids of those who are the subjec[...] Tubercular Infection—3. Distribution of Tuberculotropic Substances in [...] Infected Organism—4. Question as to whether the Protective Substances w[...] come into consideration with Tubercle are present in the Blood of the Infan[...] Birth—5. On some Points in connexion with the Elaboration by the Hu[...] Organism of Tuberculotropic Elements in response to Inoculations of a Tube[...] Vaccine—Principle upon which the Patients were Selected and General Proce[...] followed in Connexion with the Inoculations—Data furnished by the Meas[...]ment of the Agglutinating Power in the Case of Patients undergoing Anti-Tube[...] Inoculation—Data furnished by the Measurement of the Opsonic Power of Blood in the case of Patients undergoing Anti-tubercle Inoculations.

We propose to consider in this communication (1) the action exerted u[...] the tubercle bacillus by normal human blood fluids and the tuberculo[...]pic[2] substances which come here into consideration; (2) the action exer[...] upon the tubercle bacillus by the blood fluids of those who are the sub[...] of tubercular infection; (3) the distribution of tuberculotropic substan[...] in the infected organism; (4) the question as to whether these protect[...] substances are present in the blood of the infant at birth; and (5) so[...] points in connexion with the elaboration in the human organism of tub[...]culotropic substances in response to inoculations of a tubercle vacc[...]

I.—On the Tuberculotropic Elements of Human Blood and on the Cont[...] of the Normal Blood in these Elements.

Agglutinins.—The technical difficulties created by the circumsta[...]

[1] Reprinted from the *Proceedings of the Royal Society*, vol. lxxiv, 1904.

[2] The term *tuberculotropic* is, in accordance with the scheme of terminol[...] introduced by Ehrlich, employed by us to connote the property of *turning tow[...]* and entering into chemical combination with the tubercle bacillus.

the tubercle bacillus grows in artificial culture in agglomerated ses stood for a long time in the way of the demonstration of the pres- of agglutinins in the serum. These difficulties were for the first time come by Arloing, who obtained, by the operation of a process of ction, a strain of tubercle which gives a homogeneous growth when culture is frequently shaken up. For the homogeneous cultures of ing, Koch substituted a homogeneous suspension of bacillary frag- its obtained by the trituration of ordinary tubercle cultures. Koch le his suspension with physiological salt solution.

The test fluid thus constituted exhibits—and this point did not escape observation of Koch—a proneness to spontaneous agglutination. s defect, and it is a defect which may invalidate the results of any test mination, can, as was pointed out by one of us,[1] be eliminated by em- ying in lieu of the 0·85 per cent. NaCl solution prescribed by Koch, a per cent. NaCl solution.[2]

We have in the case of the investigations on agglutination which are bodied in this paper, in every case employed this 0·1 per cent. salt ition both for the dilution of the serum and for the suspension of the ercle powder.

In some of our more recent experiments we have substituted for the fluid constituted as above a homogeneous suspension of tubercle illi obtained by heating an ordinary tubercle culture to 60° C. for an r, filtering off the bacterial growth, breaking up this last in a mortar h a solution of 0·1 per cent. NaCl in 0·5 per cent. carbolic acid, and lly centrifugalizing to remove any bacterial masses which have not n resolved into their elements.

In experiments conducted with either the one or the other of these test ds agglutination effects are obtained with normal human serum. iducting the experiments in throttled capillary tubes by the method cribed by one of us,[3] and taking cognisance of the effect by naked-eye pection, a complete sedimentation is generally obtained in the two- and r-fold dilution, and incomplete sedimentation in the eight-fold ition. With some normal bloods complete sedimentation is obtained to the 16-fold dilution.

Bactericidal Elements.—Our investigations into the question as to the sence of a bactericidal element in human serum are as yet incomplete.

Opsonins.—As already brought out by us in previous papers,[4] the gocytic effect obtained when bacteria are introduced into the blood

[1] Wright, *Lancet*, July 25, 1903.

[2] The principle which suggested the replacement of the stronger by the weaker solution finds application, as one of us (S. R. D.) has recently elicited, also in case of plague cultures. The spontaneous agglutination which has up to the ent been a source of difficulty in measuring the agglutination effect exerted sera upon plague cultures can be completely avoided by employing a 0·1 per . solution of salt in lieu of the broth or physiological salt solution ordinarily loyed.

[3] Wright, *Lancet*, July 25, 1903.

[4] *Roy. Soc. Proc.*, vols. 72 and 73 (pp. 75–88 and 89–99 *supra*).

is dependent upon an action exerted by the blood fluids directly upon micro-organisms.

We have investigated this question also in connexion with the tube bacillus. In doing so a two-fold technical difficulty confronted us *first*, a difficulty associated with the circumstance that the tube bacillus is available in ordinary cultures only in the form of agglomera bacterial masses, and *secondly*, a difficulty associated with the circ stance that unaltered tubercle bacilli when they have been obtai in homogeneous suspension, are agglutinated by the action of both se and physiological salt solution.

The first difficulty can be surmounted by breaking up the bact masses in a mortar in a 0·1 per cent. NaCl solution, i.e. in a salt solu diluted up to the point at which it will no longer bring together by agglutinating action tubercle bacilli which have been mechanic separated.

The second difficulty can be surmounted by heating the tube culture to 100° C.[1]

We subjoin here a series of experiments (conducted before the proced last mentioned was thought out), with living tubercle bacilli suspende a 0·1 per cent. NaCl solution. It will be seen that the difference betw the phagocytic effect obtained with the unheated and the heated se respectively is sufficiently pronounced to throw altogether into background the source of disturbance which is associated with presence of an agglutinating element in the serum.

Preliminary Experiments.

In this series of experiments a homogeneous suspension of living tubercle ba was made by rubbing up a small quantity of a tubercle growth (obtained from a glyce potato culture) in an agate mortar in 1 in 1000 NaCl solution, and then centrifugali to get rid of the bacterial masses which had not been resolved into their elements.

Experiment 1.

A.

A. E. W.'s unheated serum	2 vols.
S. R. D.'s washed corpuscles	2 „
Suspension of living tubercle bacilli	1 vol.

Phagocytic count (average of 67 P.W.B.C.), **5·4.**

B.

A. E. W.'s serum heated to 60° C. for 20 mins. . .	2 vols.
S. R. D.'s washed corpuscles	2 „
Suspension of living tubercle bacilli	1 vol.

Phagocytic count (average of 30 P.W.B.C.), **0·75.**

Experiment 2.

A.

S. R. D.'s unheated serum	2 vols.
S. R. D.'s washed corpuscles	2 „
Suspension of living tubercle bacilli	1 vol.

Phagocytic count (average of 16 P.W.B.C.), **17·3.**

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[1] *Such heating, destroying as it does the agglutinability of the tubercle baci makes it, in point of fact, quite unnessary to employ salt solutions of low concentrat*

B.

S. R. D.'s serum heated to 60° C. for 20 mins. . . 2 vols.
S. R. D.'s washed corpuscles . . . . . 2 ,,
Suspension of living tubercle bacilli . . . . 1 vol.
Phagocytic count (average of 37 P.W.B.C.), 3·0.

Experiment 3.

A.

C. J.'s unheated serum . . . . . . 2 vols.
S. R. D.'s washed corpuscles . . . . . 2 ,,
Suspension of living tubercle bacilli . . . . 1 vol.
Phagocytic count (average of 19 P.W.B.C.), 14.

B.

C. J.'s serum heated to 60° C. for 20 mins. . . 2 vols.
S. R. D.'s washed corpuscles . . . . . 2 ,,
Suspension of living tubercle bacilli . . . . 1 vol.
Phagocytic count (average of 37 P.W.B.C.), 3·0.

/ith cultures which have been exposed to a temperature of 100° C. sely similar results are obtained, while an advantage is gained in espect that the count is no longer rendered difficult by the massing her of the bacilli.

ll the experiments hereafter subjoined have been carried out with a culture, i.e., a culture heated to 100°, broken up in 1 in 1000 solution, and centrifugalized until all unresolved clumps had been ed down.

ur next series of experiments was undertaken with a view to deterng whether the increased phagocytic effect obtained with the uned serum is due to an action exerted by the serum directly upon the rcle bacilli.

Experiment 1.

A.

S. R. D.'s unheated serum . . . . . 2 vols.
A. E. W.'s washed corpuscles . . . . . 2 ,,
Suspension of heated tubercle bacilli . . . . 1 vol.
Phagocytic count (average of 20 P.W.B.C.), 6·9.

B.

S. R. D.'s unheated serum . . . . . 2 vols.
Suspension of heated tubercle bacilli . . . . 1 vol.

he above were digested together for 15 mins. at 37° C.; were then ed to 60° C. for 10 mins.; and finally 3 vols. of the mixture were d to—

A. E. W.'s washed corpuscles . . . . . 2 vols.
Phagocytic count (average of 31 P.W.B.C.), 3·5.

C.

S. R. D.'s unheated serum . . . . . 2 vols.
Suspension of heated tubercle bacilli . . . . 1 vol.

The above were immediately, after mixture, heated to 60° C. 10 mins., and were then added to—

A. E. W.'s washed corpuscles . . . . . . 2 vols.
Phagocytic count (average of 50 P.W.B.C.), **0·16**.

Experiment 2.

A.

A. E. W.'s unheated serum . . . . . . 2 vols.
A. E. W.'s washed corpuscles . . . . . . 2 „
Suspension of heated tubercle bacilli . . . . . 1 vol.
Phagocytic count (average of 49 P.W.B.C.), **5·2**.

B.

A. E. W.'s unheated serum . . . . . . 2 vols.
Suspension of heated tubercle bacilli . . . 2 „

The above were digested together for 15 mins. at 37° C.; were heated to 60° C. for 10 mins.; and finally 3 vols. of the mixture added to—

A. E. W.'s washed corpuscles . . . . . . 2 vols.
Phagocytic count (average of 40 P.W.B.C.), **2·6**.

C.

A. E. W.'s unheated serum . . . . . . 2 vols.
Suspension of heated tubercle bacilli . . . 1 vol.

The above were immediately after mixture heated to 60° C. fo mins.; and were then added to—

A. E. W.'s washed corpuscles . . . . 2 vols.
Phagocytic count (average of 50 P.W.B.C.), **0·34**.

Experiment 3.

A.

H. B. S.'s unheated serum . . . . . . 2 vols.
A. E. W.'s washed corpuscles . . . . . . 2 „
Suspension of heated tubercle bacilli . . . 1 vol.
Phagocytic count (average of 20 P.W.B.C.), **4·8**.

B.

H. B. S.'s unheated serum . . . . . . 2 vols.
Suspension of heated tubercle bacilli . . . . 1 vol.

The above were digested together for 15 mins. at 37° C.; were heated to 60° C. for 10 mins.; and finally 3 vols of the mixture added to—

A. E. W.'s washed corpuscles . . . . . . 2 vols.
Phagocytic count (average of 30 P.W.B.C.), **2·6**.

C.

H. B. S.'s unheated serum . . . . . . 2 vols.
Suspension of heated tubercle bacilli . . . 1 vol.

The above were immediately after mixture heated together fo mins. to 60° C.; and were then added to—

A. E. W.'s washed corpuscles . . . . . . 2 vols.
Phagocytic count (average of 20 P.W.B.C.), **0·4**.

It will be manifest that these experiments testify to an opsonic a exerted by the serum directly upon the tubercle bacilli.

he smaller phagocytic effect recorded in each experiment in B as ared with A is at present without explanation.

he experiment next subjoined indicates that it is the potency of the n rather than the potency of the white corpuscles which determines mount of phagocytosis. In this experiment the corpuscles of the rcular patient's blood, and the corpuscles of the normal blood respecy were employed in A′ and A″ in combination with their native blood In B′ and B″ a reciprocal exchange of blood fluids was made.

A′.

Tubercular patient's washed corpuscles . . .	2 vols.
Tubercular patient's serum.	2 „
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 36 P.W.B.C.),	0·66.

A″.

A. E. W.'s washed corpuscles	2 vols.
A. E. W.'s serum	2 „
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 32 P.W.B.C.),	3·1.

B′.

Tubercular patient's washed corpuscles . . .	2 vols.
A. E. W.'s serum	2 „
Suspension of living tubercle bacilli	1 vol.
Phagocytic count (average of 31 P.W.B.C.),	2·1.

B″.

A. E. W.'s washed corpuscles	2 vols.
Tubercular patient's serum	2 „
Suspension of living tubercle bacilli	1 vol.
Phagocytic count (average of 30 P.W.B.C.),	1·3.

will be seen that the phagocytic effect obtained with the patient's corpuscles (in A′) was (in B′) increased more than three-fold in quence of the replacement of their native serum by that of the conblood. The phagocytic effect obtained with the white corpuscles e control blood (in A″) was (in B″) diminished in an almost corredling degree (approximately two and a half times), by the replacement eir native serum by that of the patient.

hese results are, it may be pointed out, in conformity with those ded in our previous paper [1] in connexion with the phagocytosis e staphylococcus pyogenes.

ction exerted upon the Tubercle Bacillus by the Blood-fluids of those who are the subject of Tubercular Infection.

he blood fluids of the subjects of a particular bacterial infection may xpected to differ with respect to their content in bacteriotropic ances from the blood fluids of normal persons. An increased nt in these elements may be expected in the case where there has active response on the part of the machinery of immunisation to the lus of infection; diminished content (*a*) where that machinery is ning exhausted, and (*b*) where infection is dependent upon a native, any rate, antecedent deficiency in protective substances.

[1] *Roy. Soc. Proc.*, vol. 73 (pp. 89–90 *supra*).

Agglutinating effect.—We have in no instance found the agglutina power of the blood of tubercular patients higher than that of cer normal bloods. Sometimes we have found it notably decreased. three cases we have found it altogether absent. On the average find that it does not differ sensibly from that of the normal blood

It would seem to follow—and this conclusion is in conformity that arrived at by others—that no indication as to the presence or sence of tubercular infection can be drawn from the measurement of agglutinating power, unless perhaps in the case where that reactic found to be quite absent.

Opsonic effect.—The measurement of the opsonic power of the b fluids discloses very definite differences. We have not in any of the joined patients recorded at the outset an opsonic power equal to of our own bloods. On the contrary there has been in each such ca definite defect of opsonic power. The results of our observations on point are embodied in the table below.

TABLE I.—*Showing the Opsonic Power of the Blood Fluids in a Seri Tubercular Patients.*

The procedure adopted was to mix together in each case the patient's serum with a pension of heated tubercle bacilli and with washed corpuscles derived from a n person. In each case the opsonic power of the normal serum employed as a c (derived in practically every case from A. E. W. or S. R. D.) was taken as u

Serial Number of the Observation.	Form of the Tubercular Infection.	Opsonic Inde
Case 1	Tubercular peritonitis	0.67
„ 2	Laryngeal phthisis	0·6
„ 3	Psoas abscess	0·4
„ 4[1]	Tubercular sycosis	0·4
„ 5	Nasal and pharyngeal lupus	0·56
„ 6	Tubercular cystitis	0·8
„ 7	„ „	0·6
„ 8	Generalized lupus	0·5
„ 9	Lupus of hand	0·9
„ 10	Tubercular prostatitis	0.8
„ 11	„ glands	0·85
„ 12	„ abscess of thigh	0·64
„ 13[2]	Lichen scrophulosorum	0·56
„ 14	Pulmonary phthisis	0·75
„ 15	„ „	0·69
„ 16[3]	Tubercular infection of ovary and peritonitis	0·65
„ 17	„ abscesses of arm and chest wall	0·56

[1] It is interesting to note in connexion with this case that the definite dia of tubercular infection, which was based upon the histological structure of a of excised tissue, was anticipated in view of the inductions obtained from the p cytic test recorded above.

The alternative diagnosis of staphylococcus infection was excluded by th that the patient was found to possess a normal opsonic index with respect staphylococcus.

[2] The patient gave a typical local and general reaction when inoculated fo gnostic purposes with a test dose of Koch's old tuberculin.

[3] The diagnosis was based upon a histological examination of the exti ovary.

s indicated above, a diminished content in bacteriotropic substances as we have here on record may be ascribed either to the exhaustion ie protective elements under the influence of the bacterial invasion; lternatively, to an antecedent deficiency in these elements.

The following considerations appear to us here to plead in favour of rpreting the low opsonic power of the tubercular patients here in tion as the occasion and not the consequence of infection.

a) Very low phagocytic indices have been obtained where constitu-al symptoms were absent or insignificant. The cases denoted by the l numbers 3, 4, 5, and 8 are instances in point.

b) We have in practically every case found it possible to increase, by noculation of a tubercle vaccine, the opsonic power of a patient's d fluids.

Distribution of Tuberculotropic Substances in the Infected Organism.

We have made the subjoined observations on this question. It will een that they are in consonance with the observations (see pp. 104– we have made in connexion with the distribution of the staphylococcic nins in the infected organism, and with the induction that the bac-tropic pressure is always reduced in the actual foci of infection.

Observation 1.

The patient was a child æt. 2 years, affected with tubercular necrosis ie sternum, and with a tubercular abscess in the thigh.

A.

Serum obtained from blood drawn from finger . .	1 vol.
Physiological salt solution	1 ,,
S. R. D.'s washed corpuscles	2 vols.
Suspension of heated tubercle bacilli	1 vol.

Phagocytic count (average of 20 P.W.B.C.), 3·9.

B.

Supernatant fluid of pus withdrawn from abscess .	1 vol.
Physiological salt solution	1 ,,
S. R. D.'s washed corpuscles	2 vols.
Suspension of heated tubercle bacilli	1 vol.

Phagocytic co unt (average of 20 P.W.B.C.), 1·1.

Observation 2.

The patient was a young man suffering from a psoas abscess due to ercular infection.

A.

Serum obtained from blood withdrawn from finger .	1 vol.
Physiological salt solution	1 ,,
A. E. W.'s washed corpuscles	2 vols.
Suspension of heated tubercle bacilli	2 ,,

Phagocytic count (average of 40 P.W.B.C.), 0·6.

B.

Supernatant fluid of pus derived from sinus	1 vol.
Physiological salt solution	1 „
A. E. W.'s washed corpuscles	2 vols.
Suspension of heated tubercle bacilli	2 „

Phagocytic count (40 P.W.B.C. searched), 0.

With a view to ascertaining whether the tissue lymph might r normally be poorer in opsonic substances than the serum, the follow experiment was made :—

A.

Serum of blood withdrawn from A. E. W.'s finger	2 vols.
A. E. W.'s washed corpuscles	2 „
Suspension of heated tubercle bacilli	1 vol.

Phagocytic count (average of 50 P.W.B.C.), 0·92.

B.

Fluid from blister raised by friction upon A. E. W.'s finger	2 vols.
A. E. W.'s washed corpuscles	2 „
Suspension of heated tubercle bacilli	1 vol.

Phagocytic count (average of 50 P.W.B.C.), 0·86.

Observation 3.

The patient was a young man operated upon for ascites depend upon extensive tubercular infection of the peritoneum.

A.

Serum obtained from blood withdrawn from finger	1 vol.
B. H. S.'s washed corpuscles	1 „
Suspension of heated tubercle bacilli	1 „

Phagocytic count (average of 21 P.W.B.C.) 25·4.

B.

Fluid withdrawn from peritoneum	1 vol.
B. H. S.'s washed corpuscles	1 „
Suspension of heated tubercle bacilli	1 „

Phagocytic count (average of 34 P.W.B.C.), 4·6.

It is interesting to bring into relation with the data of this last obse tion (*a*) the fact that the phagocytic count of this patient's serum stoo the phagocytic count of the control serum employed (A.E.W.'s) as 1 ; (*b*) the fact that the prognosis, so far as the restriction of the infectio the existing focus of disease, is comparatively favourable in cases of tu cular peritonitis ; and (*c*) the fact that a retrogression of the infec often follows in these cases upon the evacuation of the peritoneal fluic

The *first* and *second* of these facts suggest that a reaction of immu tion may be set up by the absorption of vaccinating elements from infected peritoneum. The *third* fact, and the same applies (*vide* Ca of the foregoing paper, pp. 104 and 105 *in this volume*), also in conne with the evacuation of abscesses may, perhaps, find its explanatio the data given above. It would be reasonable to expect that the flo new and active lymph, which would follow upon the evacuation of stagnant and exhausted lymph, would operate in the direction of ch ing the growth of invading micro-organisms.

Question as to whether the Protective Substances which come into consideration in connexion with Tubercle are present in the Blood of the Infant at Birth.

In view of the asserted superior susceptibility of infants to tubercular ction, it appeared to us to be of interest to measure the respective nic power of mother and infant. We employed for this purpose blood n from the umbilical cord and blood taken from the mother's finger ediately after completion of labour. Our observations are subjoined.

Observation 1.

A.

Serum from mother No. 1	2 vols.
A. E. W.'s washed corpuscles	2 ,,
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 45 P.W.B.C.),	1·6.

B.

Serum of infant No. 1	2 vols.
A. E. W.'s washed corpuscles	2 ,,
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 50 P.W.B.C.),	1·38.

Observation 2.

A.

Serum of mother No. 2	2 vols.
A. E. W.'s washed corpuscles	2 ,,
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 50 P.W.B.C.),	0·4.

B.

Serum of infant No. 2	2 vols.
A. E. W.'s washed corpuscles	2 ,,
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 100 P.W.B.C.),	0·37.

Observation 3.

A.

Serum of mother No. 3	2 vols.
J. F.'s washed corpuscles	2 ,,
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 40 P.W.B.C.),	3·55.

B.

Serum of child No. 3	2 vols.
J. F.'s washed corpuscles	2 ,,
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 40 P.W.B.C.),	1·85.

Observation 4.

A.

Serum of mother No. 4	2 vols.
A. E. W.'s washed corpuscles	2 ,,
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 31 P.W.B.C.),	17·9.

B.

Serum of infant No. 4	2 vols.
A. E. W.'s washed corpuscles	2 ,,
Suspension of heated tubercle bacilli	1 vol.
Phagocytic count (average of 30 P.W.B.C.),	10.

Observation 5.

A.

Serum of mother No. 4 agglutinates a suspension of fragments of tubercle bacilli completely in dilutions of 1 in 2, 4, 8, and 16; incompletely in a dilution of 1 in 32.

B.

Serum of infant No. 4 agglutinates the same suspension completely in dilutions of 1 in 2 and 1 in 4; incompletely in dilutions of 1 in 8 and 1 in 16.

5. On some Points in connexion with the Elaboration by the Hum Organism of Tuberculotropic Elements in response to Inoculatic of a Tubercle Vaccine.

We propose to set down here in briefest outline the more importa of the facts which have emerged in the course of a study of the blo changes elicited by inoculations of a tubercle vaccine undertaken therapeutic purposes.

Nature of the Tubercle Vaccine employed.—A tubercle vaccine may defined, with respect to its derivation and its effect upon the organis as any derivative of the protoplasm of the tubercle bacillus, which capable of inducing an elaboration of tuberculotropic substances in t organism.

We may include under this definition :—

(1) Such a vaccine as would be arrived at by (*a*) sterilizing a tuber culture at 60° C.; (*b*) breaking up the culture in a mortar in 0·1 per ce salt solution; (*c*) centrifugalizing to remove any residual bacterial mass (*d*) re-sterilizing at 60° C.; and (*e*) standardizing by enumeration, or centrifugalization in graduated tubes in a sufficiently concentrated s solution.

(2) The preparation which is sold as a therapeutic agent, under t name of Koch's new tuberculin or T.R. tuberculin.

This preparation consists, as is well known, of a fine suspension triturated tubercle bacilli. The trituration to which the tubercle cultu is subjected is employed with the two-fold object of sterilizing the vacc by a process of comminution, and of obtaining the fine suspension wh is desired.

It is doubtful whether the first of these ends can be efficiently secu by any process of trituration. The homogeneous suspension wh is desired can, as was shown above (p. 113), be obtained by means ot than the comminution of the bacilli by machinery.

(3) The preparation which is now sold, chiefly for diagnostic uses, ler the name of Koch's old tuberculin.[1]

This preparation consists, as is well known, of the inspissated filtrate a tubercle culture which has been grown for a period of weeks upon cerinated broth, and which has afterwards been sterilized at 100° C.

Pending the working out of a vaccine upon the lines indicated in (1), the R. tuberculin has been the vaccinating material employed.

In our earlier experiments this preparation was simply diluted with rilized salt solution.

In our later experiments we have—after satisfying ourselves that the ccinal properties of Koch's preparation are unaffected by the adoption of h precautions—in every case heated the T.R. tuberculin to 60° C. one hour, and have made our dilutions with a sterilized salt solution ich had received an addition of 0·25 per cent. lysol.

nciple upon which the Patients were Selected and General Procedure Followed in Connexion with the Inoculations.

We have in our selection of cases been guided by the desire to deal at st only with the most aggravated and seemingly intractable cases of alized tubercular infection, and only with cases which would furnish ambiguous objective evidence of any progress or regress of the ection.

The general procedure followed was to begin in each case after the asurement of the content of the patient's blood in tuberculotropic bstances, with very small doses (generally $\frac{1}{500}$ milligramme[2]) of e vaccinating material, and to reinoculate at intervals of ten days, testing the blood on each occasion, and in the case of each patient pressing the results in the form of a curve.

In our earlier experiments, before we had elaborated the procedure measuring the opsonic power of the blood, we were necessarily re-icted to a measurement of the agglutinating power.

[1] The proposition that the old tuberculin may appropriately be denoted a ccine derives its justification, first, from the consideration that the prolonged ltivation and the prolonged digestion of the culture which is involved in the ocess of manufacture must be associated with autolysis, and secondly, from e observations made in connexion with Case 13 of Table I, and the last patient Table III.

In the former case, the opsonic index of the patient's blood stood at 0·56 immedi-ely before the inoculation of 1 milligramme old tuberculin. It stood at 0·55 hours afterwards in the height of the febrile reaction, and at 1·01 three days later.

In the latter case, the opsonic index of the blood stood at 0·67 immediately fore the inoculation of 1 milligramme of the old tuberculin. It stood next day 0·4, and eight days later at 0·76.

[2] *Owing to the fact that the question of dry tubercle powder in Koch's T.R. tuber-lin was, owing to the fact that* 10 *mgr. of the powder were used for the manufacture* 1 *cubic centimetre, declared as* 10 *mgr. in* 1 *cc., when the actual content was, in reality, ly* 2 *milligrammes in* 1 *cc.* (*Ruppel, "Deutsche medic. Wochenschr.,"* 1908, *Jan.* 30), *e dose here in question would in reality be not* $\frac{1}{500}$ *milligramme but* $\frac{1}{2500}$ *milligramme.*

Data Furnished by the Measurement of the Agglutinating Power in th Case of Patients undergoing Anti-Tubercle Inoculation.

The method of investigation here in question—and we would no that it had before us been employed by Koch in connexion with inoc lations of his T.R. tuberculin—furnishes, it seems to us, indicatio which have a certain value.

An increase in the agglutinating power of the blood is generally, as the cases tabulated below, obtained in the course of a successful immu sation.

It is, however, to be noted that the rise in the agglutination cur may occur long subsequent to the achievement of very marked clinic improvement, and further that such clinical improvement may be obtain quite apart from any sensible increase in the agglutinating power of t blood.

In addition to furnishing indications of successful advance in the dire tion of immunisation, the measurement of the agglutinating power of t patient's blood may afford also indications of regress in the direction increased susceptibility resulting from an overtaxing of the machinery immunisation.

In exemplification of this we may quote three passages from the hi tory of a patient (E. S., Tables II and III) with tubercular infection of t kidney and bladder, whose agglutination curve was followed for near 18 months.

The patient in question, who had in association with the inoculatio set forth in Table II put on 5 lb. in weight, received on April 30, the da on which the record in Table II closes, 0·025[1] milligramme, on May 0·05 milligramme,[1] and on May 13 0·2[1] milligramme of T.R. tuberculin.

In association with the first two of these inoculations, the agglutin tion curve sank away rapidly from 64 to 8, the patient losing at the san time 3½ lb. in weight, and suffering from considerable constitution disturbance (see *infra*, Chart 1, fig. 271).

In association with the third of these inoculations the agglutini disappeared entirely from the blood.

On a later occasion, in the beginning of November, 1903, when t general condition of the patient had very markedly improved, and whe her body weight had increased by 23 lb., a similar negative phase effe accompanied by constitutional disturbance, was obtained in associati with the inoculation of three 1 milligramme[1] doses of the T.R. tubercul on November 2, 6, and 11 respectively. Here the complete agglutin tion, which was on the first of these dates obtained in a 32-fold diluti of the serum, was obtained after the inoculations only in an 8-fold dilutio

Again, in the beginning of December, when another attempt was ma to press the inoculations, the agglutination curve, which had risen aga to 32 after the inoculations referred to in the preceding paragraph, d clined in consequence of two 1 milligramme inoculations, first to 8 an then to 2, and the patient's symptoms were aggravated.

[1] *Vide* p. 123, *note* 2.

A similar decline of the agglutination curve has come under observa- also in other cases in association with the premature increase of the of vaccine, and with the shortening of the interval between succes- inoculations.

LE II.—*Showing the Agglutinating Power of the Blood in the case of a Series of Patients, before Inoculation, and after a Series of Inoculations of Tubercle Vaccine.*

atient's Initials.	Highest Dilution of Serum in which complete Agglutination was obtained, and Date of Observation (in brackets).	Highest Dilution of Serum in which complete Agglutination was obtained after the Inoculations particularized in the next column, and Date of Observation (in brackets).	Doses of T.R. Tuberculin, and Dates of Inoculations (in brackets).
A. R.	3 (2.12.03)	32 (23.3.04)	mgrm.[1] 0·0025 (2.12.03) 0·01 (17.12.03) 0·01 (4.1.04) 0·01 (14.1.04) 0·0075 (21.1.04) 0·0075 (1.2.04) 0·015 (15.2.04) 0·02 (1.3.04) 0·04 (11.3.04)
J. A.	0 (6.4.03)	64 (21.4.03)	0·002 (6.4.03) 0·01 (8.4.03) 0·02 (15.4.03)
N. T.	4 (6.1.04)	64 (23.3.04)	0·003 (20.1.04) 0·005 (2.2.04) 0·01 (23.2.04) 0·015 (2.3.04) 0·02 (12.3.04)
N. W.	8 (10.12.03)	32 (20.3.04)	0·01 (17.12.03) 0·0075 (4.1.04) 0·0075 (19.1.04) 0·01 (28.1.04) 0·01 (10.2.04) 0·015 (19.2.04) 0·02 (3.3.04) 0·02 (14.3.04)
E. J.	4 (27.1.04)	32 (16.3.04)	0·003 (27.12.03) 0·0066 (11.1.04) 0·01 (23.1.00) 0·015 (1.3.04)
E. S.	2 (17.4.03)	64 (30.4.03)	0·005 (21.4.03) 0·01 (24.4.03)
M. O.	0 (15.12.03)	24 (17.2.04)	0·004 (23.12.03) 0·008 (5.1.04) 0·016 (21.1.04)

The weights here in question refer in each case to the weight of tubercle er stated to be contained in the quantum of T.R. tuberculin administered. t *vide* p. 123, *note* 2).

TABLE III.—*Shows the Changes induced in the Opsonic Power of the Blood Fluids by the Inoculation of Tubercle Vaccine and furnishes Illustration of the Fact that the Cumulative Increase of the Protective Elements which is desired can be achieved only by the Proper Regulation and Interspacing of the Successive Doses of Vaccine.*

Initials.	Brief History of Case.	Dates and Particulars of Inoculations with T.R. undertaken since March, 1904.		Opsonic Index (Opsonic Power of the normal Blood —A. E. W.'s— used as a Control was taken as = 1).
E. J. . .	Patient, a man of 30, developed tubercular glands on the left side of neck and a tubercular abscess on the point of the shoulder in the autumn of 1902. He was admitted to hospital and was operated upon for the first time in January, 1903. The wounds becoming invaded with tubercle, and refusing to heal, while the area of infection gradually extended, six further (scraping, extirpating, and skin-grafting) operations were undertaken during the course of the year. In December, 1903, when patient came up for treatment by inoculation, the whole area from the point of the shoulder to the ear had been converted into an ulcerated surface, there was a deep crater undermining the angle of the jaw and the ear, the left side of the face was distorted with swelling, and the axilla was occupied by a gland as large as a pigeon's egg. The patient was very anaemic and emaciated. Steady improvement has been made under the inoculation treatment, the swelling of the face has almost entirely dispersed, the cavity under the jaw has healed up from the bottom, the gland in the axilla can no longer be felt, the ulcerated wound surfaces have almost completely closed, and the patient has the constitutional aspect of a healthy man.	 28.3.04 18.4.04 25.4.04 10.5.04 7.6.04 27.6.04 15.7.04 19.7.04	mgrm.[1] 0.04 0·1 0·1 0·1 0·1 nil 0·02 nil	 — 0·73 — — 1·6 1·45 — 1·15

Note.—During the period 7.6.04–15.7.04 the inoculations of tubercle vaccine were suspended while the patient was being immunised against the staphylococcus pyogenes, which, by its presence on wound, appeared to be preventing the process of healing.

	constantly present in phenomenal numbers in the urine, and a tubercular ulcer of the bladder was detected. Patient had been losing flesh and suffered from night sweats and from constant pain and frequency.	27.4.04	nil	0·42
	Treatment by inoculation was begun on 21.4.03 and has been continued up to date, inoculations and examinations of the blood and urine being undertaken on an average once in 10 days.	9.5.04	0·5	0·77
	Under the treatment the tubercle bacilli in the urine have gradually diminished, and since the beginning of May they have been completely absent. The patient has increased 36 lb. in weight, and is now to all intents and purposes well, except for an infection of the urinary tract by the bacillus coli and pneumococcus, which is being dealt with by the inoculation of the appropriate vaccines.	1.6.04	0·5	0·4
		15.6.04	nil	1·3
		6.7.04	nil	2·2

Note.—The results of the blood examinations bring out that the inoculations were here conducted with excessive doses or perhaps at too short intervals. It will be seen that the opsonic power of the patient's blood declined in consequence of the inoculation undertaken on 8.4.04 from 0·93 on that date to 0·4 on 18.4.04. On the date last mentioned another 0·5 milligramme was inoculated—and it may be noted in this connexion that here as elsewhere it was often necessary to inoculate before the result of the blood examination was known. On 27.4.04, recovery was still incomplete, the patient being obviously poorly, and the inoculation was postponed. When next examined twelve days later the opsonic power of the blood was found to have risen to 0·77. A further dose of 0·5 milligramme was then inoculated, with the result that the opsonic power was reduced again to 0·4 on 1.6.04. On this day the patient again received a dose of 0·5 milligramme, the effect of this became manifest fourteen days after in the rise of the opsonic power to 1·3 and thirty-six days after in the rise of opsonic power to 2·2.

N. W.	Patient, a woman 31 years of age, developed a tubercular infection of the glands of the neck at the age of 14.	18.4.04	0·04	0·16
	When the abscesses which formed in association with these were opened the overlying skin became invaded, the infection spread to other glands, and tubercular disease developed in the little finger of the right hand. The two terminal joints of this finger were removed when the patient was 16.	3.5.04	0·05	0·6
	When the patient was 19 she was treated with Koch's old tuberculin, receiving three to four inoculations a day (150 inoculations in all). As a result of these inoculations the patches of lupus on hand, neck and face became inflamed, a piece of bone sloughed out of the arm and the patient lost weight and became seriously ill. She remained in hospital in all thirteen weeks.	16.5.04	0·05	—
		1.6.04	0·05	0·78
		10.6.04	nil	—
		19.6.04	0·04	—
		6.7.04	0·04	1·85
		22.7.04	—	1·7

[1] The weights here in question refer in each case to the weight of tubercle powder stated to be contained in the quantum of T.R. tuberculin administered. (*Vide* p. 103 *note* 2.)

TABLE III—*continued.*

Initials.	Brief History of Case.	Dates and Particulars of Inoculations with T.R. undertaken since March, 1904.		Opsonic Index (Opsonic Power of the normal Blood —A. E. W.'s— used as a Control was taken as = 1).
			mgrm.	
	Four years later energetic local treatment was adopted, and scraping operations were undertaken upon the glands in the neck. In 1900 the Finsen light treatment was adopted, and was persevered in for eighteen months. This effected an improvement in the condition of the face and neck, but the disease continued to extend in the deeper structures, and in particular in the bones of the left arm. Finally it became necessary to amputate this limb. The disease now broke out in the stump, on the point of the shoulder, and in the chest wall, while it persisted all over both sides of the face and neck. After Röntgen rays had been tried unavailingly, recourse was had to inoculations of tubercle vaccine, the treatment being begun on December 10, 1903. After six months' treatment the discharge from the stump and chest wall has practically ceased, the patch on the point of the shoulder has healed up, the face appears to be in a better condition, and the patient's general health, which was previously very unsatisfactory, has improved in a remarkable manner. Her body weight has steadily gone up, and has now reached 141½ lb., as much as 5 lb. having on one occasion been gained in the interval between two successive inoculations.			
A. R. .	Patient, when referred for treatment by antitubercular inoculations in December, 1903, was found to be an emaciated, anaemic, physically and mentally undeveloped child of 19 affected by lupus of the nose, throat, angle of jaw, and feet and hands. The last were a mass of ulceration, the bones of the hand being involved in many places. After six months' teatment the patches of lupus on the nose and throat have almost dried up, and the condition of the hands and feet is much amended	22.3.04	0·02	—
		7.4.04	0·05	1·1
		18.4.04	0·1	0·25
		28·4.04	nil	—
		5.5.04	0·05	0·61
		6.5.04	nil	0·85
		16.5.04	0·1	—
		7.6.04	0·1	1·2

Note.—The obviously excessive dose of 0·1 milligramme was administered on 18.4.04 before it had been elicited by blood examination that the patient had not yet responded to the dose of 0·05 milligramme administered on 7.4.04. As a result—it may be assumed—of this cumulation of doses considerable constitutional disturbance was experienced. In view of this and of the development of a phlyctenule on the eye (a phenomenon which had once before been noticed in connexion with the development of a negative phase) the inoculation fixed for the 28.4.04 was postponed and on the next occasion a smaller dose was administered.

A. A. . .	Patient, a young man, had been in bed for twelve months with a psoas abscess which discharged continuously and showed no signs of improvement. Treatment by inoculation was begun on April 8. The patient is reported to have made rapid progress towards recovery immediately after the adoption of the treatment.	8.4.04	0·002	0·4
		20.4.04	0·002	—
		3.5.04	—	0·5
		10.5.04	0·004	—
		31.5.04	0·01	—
		2.6.04	—	0·8
		16.6.04	—	1·1
M. O. .	Patient, a young married woman, developed a double psoas abscess, which was opened up before and behind in January, 1903. The discharge from the wound continued till December without any sign of improvement, the temperature reaching 101° F. practically every night. The treatment by inoculations of tubercle vaccine was begun in the middle of December, 1903, and has been continued since that date. The temperature became normal ten days after the first inoculation, the wounds are practically healed and the patient is able to go out for drives. Her body weight has increased by 16 lb.	Record of the inoculations which were carried out by the surgeon in charge of the case is not to hand.		
		10.6.04	0·1	—
		19.6.04	—	1·95
S. T. . .	Patient, a man of 35, is the subject of phthisis associated with tubercular disease of the larynx.	2.6.04	0·002	0·6
		8.6.04	—	1·15
		9.6.04	0·004	—
		21.6.04	—	0·98
		22.6.04	0·008	—
		12.7.04	0·008	0·84
		25.7.04	—	1·36

K

[1] *Vide* p. 123, *note* 2.

TABLE III—*continued.*

Initials.	Brief History of Case.	Dates and Particulars of Inoculations undertaken since March, 1904.		Opsonic Index (Opsonic Power of the normal Blood —A. E. W.'s— used as a Control was taken as = 1).
			mgrm.[1]	
S. N. . .	Patient, a woman æt. 34, developed at age of 14 a tubercular abscess in left shoulder, at age of 16 a similar affection of the right leg behind knee, and at 17 lupus of the hand and foot. The little finger of the right hand was amputated in 1887, the third finger of the same hand in 1903, and three toes in April, 1904. In each case the tubercular infection has recurred in the site of the wound.	13.6.04	0·004	0·9
		21.6.04	0·01	—
		27.6.04	0·005	0·7
		5.6.04	—	0·65
		14.7.04	—	1·05
		15.7.04	0·008	—
		21.7.04	—	0·73
S.B. . .	Patient, a man of 22, has been the subject of lupus for the last fifteen years. He has been treated by all the ordinary methods, including the application of Röntgen rays. Practically the whole of his face and neck are affected. He has also large patches on the scalp, trunk, and elbow of the right arm.	7.7.04	0·002	0·5
		11.7·04	—	0·44
		15.7.04	0·004	—
		18.7.04	—	1·3
P. C. . .	Patient, a woman of 55, has for a long period of years been the subject of lupus of nose. The affection has now spread to her pharynx and larynx.	5.7.04	0·002	0·56
		12.7.04	0·004	0·48
		19.7.04	—	1·0
J. S. . .	Patient, a man æt. 35, has suffered for two years from an inflammatory tumefaction of the subcutaneous tissues in the region of the angle of the jaw and anterior portion of the throat. The patient has been treated by scraping and the application of antiseptics.	4.7.04	1·0[2]	0·67
		5.7.04	—	0·4
		13.7.04	0·004[3]	0·76
		21.7.04	—	1·06

[1] *Vide* p. 123, *note* 2. [2] Old tuberculin. [3] New tuberculin.

a furnished by the Measurement of the Opsonic Power of the Blood in the case of Patients undergoing Anti-tubercle Inoculations.

Much more valuable than the indications which can be gleaned from measurement of the agglutinating power of the blood are the indica-s furnished by a measurement of the opsonic power of the patient's d. While the measurement of the agglutinating power of the blood fail to furnish indications of an abnormally low resisting power on the of the untreated patient ; and while it may yield only tardy informa- of the alterations effected in the blood fluids by inoculation ; and e it may sometimes altogether fail to distinguish between the patient's lition before and after successful immunisation ; the measurement ne opsonic power satisfies, it would seem, all these desiderata.

We have already seen in Table I that it distinguishes [1] between the ercular subject and the person with normal resistance. And we shall n the table below that it furnishes prompt and clear indication of the tive phase which supervenes upon inoculation, and again of the posi- phase whch succeeds the negative phase wherever the organism esses the necessary power of response. Furthermore the measurement ne opsonic power of the blood distinguishes clearly between the un- ted tubercular patient and the patient who has made progress in the ction of immunisation. This will appear clearly on comparing, in le III below, the opsonic indices achieved after inoculation with those forth in Table I.

In conclusion we may note that while we are jointly responsible for the rvations set forth in Sections I, 3, and 4 of this paper, the work h is embodied in Sections 2 and 5 has been separately undertaken one of us.

[1] It will not, however, invariably do so.

Experiments on the Nature of the Opsoni
Action of the Blood Serum.[1]

By William Bulloch, M.D., and E. E. Atkin, B.A.

From the Laboratory of the Bacteriological Department, London Hosp
London, E.

Effect of Heat upon Serum containing Opsonin—Effect of Cold upon Opsor Effects of Exposure to Light—Are the Opsonins, or are the Leucocytes, the Var Factors when the Phagocytic Power of Different Bloods is Compared?—O Constitution of the Opsonic Body in the Serum—Can the Opsonin ac Bacteria which have been subjected to High Temperatures?—Can Prolo Heating at 60° C. of Opsonised Cocci destroy the Opsonic Power so that Cocci cannot be picked up subsequently by Leucocytes?—Experiments or Rate of Disappearance of Opsonin from Serum when the latter is brought Contact with Cocci at 37° C. and at 0° C.—Experiments to Determine Nature of the Opsonic Body, and its Mode of Action—If Heated Seru unable to exert an Opsonic Action on Staphylococci, are the latter, when dig with Heated Serum, capable of being Opsonised by Unheated Serum?

In a series of simple and convincing experiments Wright and Doug have shown that in phagocytosis so called, an important if not a card rôle is played by the body humours, whereby they act upon the bact thus rendering the latter an easy prey for the polynuclear leucoc The demonstration of this *opsonic* action of the serum or plasma mainly brought about by testing separately and combined the b humours and the corpuscles, which had been washed in salt solu Contrary to general opinion, Wright and Douglas found that the cocytes were capable of engulfing microbes only when the latter been attacked by the serum or plasma. This attack on the microbe not lead to the death of the latter, as sera may manifest a marked ops effect without being in the slightest degree bactericidal. Wright Douglas found that the opsonic substance was more or less thermola being destroyed in ten to fifteen minutes at a temperature of 60° to 65 In subsequent papers these authors have demonstrated that there definite type of immunity in which the blood fluids co-operate with leucocytes to destroy the invading micro-organisms, this being diffe from the antitoxic and bactericidal types of immunity which have alr been studied with completeness.

[1] Reprinted from the *Proceedings of the Royal Society*, vol. lxxiv, 1905.
[2] *Roy. Soc. Proc.*, vols. lxxii and lxxiii (pp. 75–99 *supra*).

Technique.—The technique we have employed is that described by ght, and for the most part the experiments were made with living ures of *Staphylococcus albus* not more than twenty-four hours old. For ırate and uniform results it is essential that the emulsions of the ures should be homogeneous, the bacteria being uniformly distributed separated from each other. This is best obtained by shaking, and the sequent application of the centrifuge. From their tendency to group nselves into masses, certain strains of staphylococci are unsuitable letermining the opsonic power of the serum. Where different bacterial ılsions are compared with each other it is essential that they should ain the same number of bacteria, a result best obtained by counting diluting as required.[1] The leucocytes were obtained from the ıted blood of the authors and other human beings or from rabbits. sera used were either of human or animal origin, and were for the t part from normal individuals.

[n all cases the proportion of serum, bacteria, and corpuscles was 1 : 3.

Effect of Heat upon Serum containing Opsonin.

Serum ceases to exert an opsonic effect upon bacteria after it has been ced in the water bath at 60° to 65° for ten to fifteen minutes. In t cases the opsonic effect is totally abolished at this temperature, in w cases, however, some slight effect can be witnessed, but this is nly, if not entirely, due to traces of serum left attached to the ocytes where these have been incompletely washed in normal solution.

Experiment.

Normal rabbit's serum (three parts), mixed with staphylococcus ılsion (one part), and washed human blood corpuscles (three parts). ortion of this was tested, the serum being unheated. Other portions heated to 60° C. for varying periods. In each case a phagocytic ıt was made by numbering the cocci in fifty leucocytes and then ing the average per leucocyte.

						Cocci per leucocyte.
1.	(Control). Normal serum		+ cocci	+ corpuscles	=	14
2.	Serum heated to 60° C. for	3′	+ ,,	+ ,,	=	0
3.	,, ,,	6′	+ ,,	+ ,,	=	0
4.	,, ,,	9′	+ ,,	+ ,,	=	0
5.	,, ,,	12′	+ ,,	+ ,,	=	0
6.	,, ,,	15′	+ ,,	+ ,,	=	0

The opsonin can, however, be destroyed at even lower temperatures ıe heat is prolonged.

[1] See Wright, *Lancet*, July 5, 1902.

Experiment.

Here the conditions of the experiment were the same, with exception that the serum was heated to 55° and 50° respectively, ins of at 60° C.

									Cocci pe leucocyte
(1)	1. (Control). Unheated serum		+	cocci	+	leucocytes	=		10·9
	2. Serum heated to 55° C. for	30′	+	,,	+	,,	=		0·3
	3. ,, ,,	60′	+	,,	+	,,	=		0
(2)	1. (Control). Unheated serum		+	cocci	+	corpuscles	=		13
	2. Serum heated to 50° C. for	10′	+	,,	+	,,	=		3·4
	3. ,, ,,	15′	+	,,	+	,,	=		2·4
	4. ,, ,,	20′	+	,,	+	,,	=		2
	5. ,, ,,	25′	+	,,	+	,,	=		1
	6. ,, ,,	30′	+	,,	+	,,	=		1

Effect of Cold upon Opsonin.

Cold exerts little effect upon the opsonic power of the serum ; w immersed in ice water for twenty-four hours the opsonic value s about one-third. At ordinary temperatures the opsonin is remark stable, showing practically no diminution for twenty-four hours.

Effects of Exposure to Light.

In ordinary diffused daylight the opsonic power of the serum rem unaltered for many hours, but when exposed to bright sunlight for t hours a serum was seen to become less opsonic in the proportion of 10

Are the Opsonins, or are the Leucocytes, the Variable Factors when Phagocytic Power of Different Bloods is Compared?

For many years Metchnikoff has taught that the leucocyte is the domi factor in phagocytosis. He has also emphasized the training of the leuco as the essential thing in immunity. The experiments we have made con the results already obtained by Wright, and point to the conclusion t the leucocyte is indifferent, the variable in a series of bloods being serum. In a first series of experiments the *leucocytes* derived from se different persons were tested with respect to their phagocytic po one and the same staphylococcic emulsion and one and the same se (rabbit's) being used in each case.

In a second series the *sera* of the seven individuals whose corpus had been used in the above experiment were tested in respect of t opsonic power, one and the same suspension of cocci, and one and same variety of leucocytes (derived from one of ourselves, W.B.) b employed throughout.

Experiment 1.

Rabbits' serum mixed with emulsion of staphylococci and human
ocytes (from seven persons) in the proportion of 3 : 1 : 3. Phagocytic
it obtained by counting the number of cocci in thirty-five polynuclear
ocytes, and then calculating the number per leucocyte.

							Cocci per leucocyte.
1.	Rabbit's serum	+	cocci	+	corpuscles	(of W. B., a normal male)	= 9·8
2.	,,	+	,,	+	,,	(of F. T. ,,)	= 9·3
3.	,,	+	,,	+	,,	(of O. G. ,,)	= 9·7
4.	,,	+	,,	+	,,	(of R. D. ,,)	= 9·6
5.	,,	+	,,	+	,,	(of C. H. ,,)	= 9
6.	,,	+	,,	+	,,	(of H. M., an anæmic female)	= 9·9
7.	,,	+	,,	+	,,	(of S. M., male, facial acne)	= 9·0

Experiment 2.

Various human sera + cocci + one kind of leucocytes from a normal
e individual.

									Cocci per leucocyte.
1.	Serum of	W. B.	+	cocci	+	corpuscles of	W. B.	=	21·3
2.	,,	F. T.	+	,,	+	,,	,,	=	20·3
3.	,,	O. G.	+	,,	+	,,	,,	=	21·1
4.	,,	R. D.	+	,,	+	,,	,,	=	20
5.	,,	C. H.	+	,,	+	,,	,,	=	19·8
6.	,,	H. M.	+	,,	+	,,	,,	=	15·5
7.	,,	S. M.	+	,,	+	,,	,,	=	14

The possible objection that the human leucocytes would be injured in
tact with rabbit's serum is disposed of by an experiment in which
phagocytic power of one serum, either human or rabbit's, was deter-
ed with both human and rabbit's leucocytes. Even in this case the
ocyte is largely an indifferent factor.

							Cocci per leucocyte.
1.	Rabbit's serum	+	staphylococci	+	rabbit's	leucocytes	= 10·3
2.	,,	+	,,	+	human	,,	= 13
3.	Human serum	+	,,	+	rabbit's	,,	= 19·5
4.	,,	+	,,	+	human	,,	= 17·6

The preponderating effect exerted by the serum as distinguished
n the polynuclear leucocytes is again brought out strikingly in the
owing experiment. In this case the serum mixed with corpuscles
a normal individual (W.B.) is compared with the sera and the
puscles of three advanced cases of facial lupus, the first of these
es (No. 2 and No. 5) being a wretchedly under-nourished girl
een with very extensive facial lupus and large ulcerated tubercular
es on the hands. The test material in this experiment was an emulsion
tubercle bacilli, and the number of T.B. in one hundred leucocytes
counted and the average struck as usual.

	T.B leucoc
1. Serum of normal individual + T. B. emulsion + leucocytes of normal individual	= 5·
2. " " + " + leucocytes of a lupus patient	5·
3. " " + " + " "	= 5·
4. " " + " + " "	= 5·
5. Serum of a lupus patient + " + leucocytes of a normal individual ..	= 2·
6. " " + " + " "	= 2·
7. " " + " + " "	= 3·

Expressing these relations as the opsonic index, we have—

No. 1 = 1·0 = (normal serum + normal leucocytes).
" 2 = 0·94 = (" + leucocytes from lupus patient).
" 3 = 0·90 = (" + " ").
" 4 = 0·93 = (" + " ").
" 5 = 0·43 = (lupus serum + normal leucocytes).
" 6 = 0·43 = (" + ").
" 7 = 0·56 = (" + ").

On the Constitution of the Opsonic Body in the Serum.

The bacterial bodies already known to exist in the serum are refera to one of three classes, viz., antitoxins, agglutinins, lysins. The last these are the most complex, as the lytic action occurs only after coalition of two distinct elements, the complement which for the m part is thermolabile, and the immune body, which is thermostal The agglutinins are of simpler constitution, although here special co ditions of the agglutinating substance (the serum) and the agglutina substance (the bacteria) must exist before agglutination is manife Temperatures above 70° C. applied either to the serum or to the bacte suffice to inhibit the agglutination, although the agglutinin is apparen not entirely destroyed. In comparison with the lysins and agglutin the antitoxins are believed to be relatively simple bodies. We ha asked ourselves the question, to which, if any, of these classes do opsonins belong, and we have followed the usual methods of expe mentation which have been utilized to determine the constitution such antibodies. These experiments chiefly consist in determining temperature at which the specific action is abolished, the temperatu at which the antibody enters into combination with the bacteria, a whether the action of heat is one of destruction or merely a conv sion into some modification in which the specific action is no long manifest.

Can the Opsonin act on Bacteria which have been subjected to Hi Temperatures?

Experiment 1.

Technique.—Emulsions of cultures of staphylococcus were plac

ealed glass tubes and subjected to temperatures of 100 to 134° C. varying periods, the opsonic action of serum on such heated cultures g compared with that upon unheated emulsions.

Results.—

			Cocci per leucocyte.
Normal human serum	+ unheated cocci	+ leucocytes	= 30
" "	+ cocci heated to 100° C. for 30′	+ "	= 20
" "	+ " " 100° C. for 60′	+ "	= 20
" "	+ " " 115° C. for 30′	+ "	= 15
" "	+ " " 120° C. for 30′	+ "	= 15

Experiment 2.

			Cocci per leucocyte.
Human serum	+ unheated cocci	+ corpuscles	= 27
"	+ cocci heated to 100° C. for ½ hr.	+ "	= 17
"	+ " " 1 "	+ "	= 15
"	+ " " 1½ hrs.	+ "	= 12·9
"	+ " " 2½ "	+ "	= 13
"	+ " 134° C. for 1½ "	+ "	= 12·6

From these experiments it is apparent that there is a certain falling-f the opsonic action when the cultures are kept at high temperatures ong periods, but even at 134° C. for 1½ hours the diminution in phago-c power is about 50 per cent. It is possible, too, that the readings really higher, as it is often very difficult to count the bacteria which e been subjected to such high temperatures on account of defective etration of the stains employed.

Effect of heating to 60° C. a mixture of serum and cocci which have ady been digested at 37° C. for fifteen minutes.

We have already seen that a temperature of 60° C. applied to the m suffices to abolish its opsonic effect. Wright and Douglas showed, ever, that if the serum were first brought into contact with bacteria 37° C. for fifteen minutes, and the mixture were then heated to C. for fifteen minutes, the cocci were picked up by leucocytes without culty. It was upon this experiment that they based their conclusion the opsonin really acts upon the bacteria and does not merely ulate the leucocyte.

Prolonged Heating at 60° C. of Opsonised Cocci destroy the Opsonic Power so that these Cocci cannot be picked up subsequently by Leucocytes?

Experiment.

Technique.—Normal serum (three parts) mixed with staphylococcus lsion (one part); mixture kept in water bath at 37° C. for fifteen ntes. This mixture, which is spoken of below as "opsonised cocci," then distributed into a series of glass pipettes which were placed in

the water bath at 60° C. for periods of fifteen minutes up to five hou
On removal from the water bath, four volumes of the "opsonised cocc
were mixed with three volumes of corpuscles at 37° C. for fifte
minutes, and the phagocytic count made as usual. For comparis
the phagocytic count of unheated serum + cocci, and of serum heat
to 60° C. before being mixed with cocci, is added.

Results.—

					Cocci per leucocyte.
1.	Unheated serum (3 vols.)		+ cocci (1 vol.)	+ corpuscles (3 vols.)	= 28
2.	Serum heated to 60° C. for 15		+ „ „	+ „ „	= 0·1
3.	"Opsonised cocci" at C. 60° for	15′ (4 vols.)		+ „ „	= 27
4.	„ „	30′ „		+ „ „	= 28
5.	„ „	45′ „		+ „ „	= 23
6.	„ „	60′ „		+ „ „	= 24·
7.	„ „	1½ hrs. „		+ „ „	= 23
8.	„ „	2 „ „		+ „ „	= 23·
9.	„ „	2½ „ „		+ „ „	= 22
10.	„ „	3 „ „		+ „ „	= 24
11.	„ „	5 „ „		+ „ „	= 23·

The experiment shows that some change is produced in the bacte
during the fifteen minutes' exposure at 37° C., and the change is su
that subsequent heating to 60° for even five hours is inoperative, t
being very different to the effect of a preliminary heating of serum
60° C. before admixture with bacterial emulsion.

**Experiments on the Rate of Disappearance of Opsonin from Serum wh
the latter is brought into contact with Cocci at 37° C. and at 0°**

Experiment.

Technique.—Normal serum mixed with an equal volume of staphy
coccus emulsion and then filled into a series of capsules. The capsu
were sealed and placed in the water bath at 37° C. or in a mixture of
and salt. After varying periods the capsules were removed and carefu
centrifugalized for one hour, the clear supernatant fluid from each caps
being tested upon a fresh suspension of staphylococcus to see wheth
the opsonin had disappeared. As a control the opsonic power of norm
serum in proper dilution is also added, likewise the opsonic power
serum which has been heated to 60° C. for fifteen minutes.

					Cocci leucoc
1.	(Control). Normal serum (3 parts)		+ cocci (1 part)	+ corpuscles (3 parts)	= 18
2.	(Control). Heated „ „		+ „ „	+ „ „	= 0
3.	Supernatant fluid from capsule at 37 C. for	5′	+ „ „	+ „ „	=
4.	„ „ „	10′	+ „ „	+ „ „	=
5.	„ „ „	15′	+ „ „	+ „ „	=
6.	„ „ „	30′	+ „ „	+ „ „	=
7.	„ „ „	45′	+ „ „	+ „ „	=
8.	„ „ at 0° C. for	10′	+ „ „	+ „ „	=
9.	„ „ „	20′	+ „ „	+ „ „	=
10.	„ „ „	30′	+ „ „	+ „ „	=
11.	„ „ „	45′	+ „ „	+ „ „	=

The result is unequivocal ; the opsonin had completely disappeared
1 the serum within ten minutes both at 37° C. and at 0° C.

eriments to Determine the Nature of the Opsonic Body, and its Mode of Action.

We have seen above, that when heated to 60° C., serum ceases to exert
opsonic effect. We have also seen that opsonin disappears from the
m when the latter is digested with bacteria at 37° C., or at 0° C. Is
disappearance due to destruction, or does the opsonin pass into
e modification, in which an opsonic effect is not visible ? Is the
nin a simple or a complex structure ?

Experiment 1.

Technique.—(1) Normal serum was digested with an equal quantity
mulsion of cocci at 37° C. for fifteen minutes. After digestion,
ture was centrifugalized for ¾ hour. In this way a clear supernatant
l (A) was separated from a deposit of cocci (A′).

2) Normal serum was digested with an equal quantity of emulsion
occi at 0° C. for fifteen minutes, centrifugalized as above, and
rated into a supernatant fluid (B) and a deposit (B′).

As the serum was mixed with equal quantity of cocci, the controls
e with normal serum and " normal " cocci are given in their appro-
te dilutions, which were made with 0·85 per cent. saline. The term
rmal " is applied to cocci which have not been treated in any way.

Results.—

			Cocci per leucocyte.
ontrol). Normal serum (2-fold dilution)	+ " normal " cocci	+ corpuscles	= 25
,, ,, (4-fold ,,)	+ ,,	+ ,,	= 17
luid A	+ ,,	+ ,,	= 1
,, B	+ ,,	+ ,,	= 2·5
,, A	+ cocci B′	+ ,,	= 27
,, A + B	+ " normal " cocci	+ ,,	= 3
ontrol). Normal serum (undiluted)	+ ,,	+ ,,	= 31
,, ,,	+ cocci A′	+ ,,	= 28
,, ,,	+ ,, B′	+ ,,	= 26·5

This experiment again shows the disappearance of the opsonin from
serum at 37° and 0° C. It also shows (Nos. 8 and 9) that the opsonin
passed into the cocci (A′ and B′).

Experiment 2.

In this experiment an attempt was made to determine whether at
. a complement-like body could be separated out, which, with heated
m, would exert an opsonic effect.

Technique.—1. Normal serum mixed with emulsion of cocci, in equal
s, at 0° C. for fifteen minutes. The mixture was then centrifugalized,
separated into a supernatant fluid (A) and a deposit (A′).

2. Serum *heated* to 60° C. for fifteen minutes, mixed with cocci ($\overline{a}$ and placed at 0° C. for fifteen minutes. It was then centrifugalized, a separated into a supernatant fluid (B), and a deposit (B′). Contr were supplied by normal serum (in appropriate dilutions), digested wi "normal" cocci and corpuscles at 37° C., and heated serum under t same conditions.

						Cocci leucoc
1.	(Control). Normal serum (2-fold dilution)	+	"normal" cocci	+	corpuscles	=
2.	„ „ (4-fold „)	+	„	+	„	=
3.	„ Serum heated to 60° C. for 15′	+	„	+	„	=
4.	Fluid A + heated serum ($\overline{aa}$)	+	„	+	„	=
5.	Fluid A	+	„	+	„	=
6.			cocci A′	+	„	=
7.			cocci B′		„	=

Remarks.—This experiment shows that the opsonin is very differe in type from a lysin, as apparently no complement-like body rema in the supernatant fluid after the serum has been digested at 0° C. T low reading in the case of the deposit B′ would also lead to the supposit that the action of heat at 60° has been to destroy the opsonin altogeth This supposition in confirmed by the following experiment.

If Heated Serum is unable to exert an Opsonic Action on Staphylococ are the latter, when digested with Heated Serum, capable of bei Opsonised by Unheated Serum?

Experiment.

Technique.—Normal serum was heated to 60° C. for fifteen minut then mixed with an equal quantity of staphylococcus emulsion, a digested at 37° C. for fifteen minutes, the deposit of cocci (A) being th removed by the centrifuge.

The deposit (A) was then divided into two parts, one being reserv for the opsonic test, the other being mixed with *normal* serum. T mixture was digested for thirty minutes; the centrifuge was then applic so as to separate a supernatant fluid (B) from a deposit of cocci (B′).

Results.—

						Cocci leucoc
1.	Control). Normal serum	+	normal cocci	+	corpuscles	= 18·6
2.	„ „ (2-fold dilution)	+	„	+	„	= 16
3.	„ Heated serum	+	„	+	„	= 0·
4.	Normal serum	+	cocci A	+	„	= 15·
5.	Fluid B	+	normal cocci	+	„	= 0·
6.			cocci B′	+	„	= 15

This experiment bears out the suggestion above, that heat destro the opsonin. The cocci (A) having been quite uninfluenced by t heated serum, were capable of being opsonized by normal serum, a they were further, as shown in No. 5, capable of abstracting all the opsor

of normal serum. After being acted on by the normal serum, the
addition of corpuscles demonstrated that they had been acted upon
he opsonin (No. 6).

esults.—

. Opsonin is present in the normal serum.

. Opsonin is thermolabile.

. It rapidly disappears from the serum when the latter is mixed
bacteria at 37° C. or at 0° C.

. After the opsonin has united with the bacteria the mixture of
n and cocci can be heated to 60° C. for long periods without abolition
ie opsonic effect.

. The leucocyte is practically an indifferent factor when the phago-
power of different bloods is compared.

. The capacity of bacterial emulsions for extracting opsonin from
serum is only slightly diminished by subjecting these emulsions to
high temperatures over prolonged periods.

. The action of heat is to destroy the opsonin, and not merely to
ert it into a non-opsonizable modification.

. The opsonin is not identical with any of the antibodies hitherto
vered in the serum.

. The opsonin is of relatively simple constitution; where these
riments cover the same ground as those of Wright and Douglas, the
rvations of these authors are confirmed.

Inquiry into the Opsonic Content of the Blood Serum in Healthy Individuals and in Patients affected by Lupus.[1]

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Determination of the Opsonic Content of Normal Individuals—Determinations o the Opsonic Index of Individuals suffering from Lupus.

The following observations were made to determine upon a fairl large scale the opsonic content of the serum of individuals suffering fro lupus in comparison with that which obtains in individuals presumabl healthy, or, at any rate, not suffering from tuberculosis. Manifestly i is a matter of importance to determine, first of all, whether there ar variations in the opsonic content of the serum in healthy individuals, a the test employed at the present time necessitates the use of serum fro a normal individual as a control.

The discoveries of Wright and Douglas of the opsonic content o the serum have created wide interest in clinical circles and a desire t apply these methods in medical practice. Past experience shows tha disappointment may follow the precipitate anxiety on the part of th clinician, and it becomes necessary to determine on as wide a basis a possible the fundamental data which underlie the phenomena discovere by Wright, and from a practical standpoint to determine on a large numbe of people, healthy and diseased, the variation which may occur in th opsonic index. It is only in this way that an opinion can be forme on the opsonic test as a diagnostic or prognostic aid. The followin observations have been carried out with this aim, the number of indivi duals examined being 216, viz. 66 normal and 150 cases of skin tuber culosis, mostly in the form of lupus, obtained from the Skin Departmen of the London Hospital. These lupus cases constitute an importan series, as they were all undergoing a system of treatment by Finsen light and it has been possible to form some opinion of the relation of the opsoni test to the results obtained by the Finsen light. The statements mad subsequently as to the cure of the particular lupus patients were in al

[1] Reprinted from the *Transactions of the Pathological Society of London*, vol. lv Part III, 1905.

ses supplied to me independently by those in charge of the Finsen ierapy Department *after* I had made the opsonic determinations.

In all cases the serum was tested within an hour or two after the blood d been withdrawn from the body, and the test material employed was . emulsion of tubercle bacilli. It cannot be sufficiently emphasized at the preparation of this emulsion is of fundamental importance, d a considerable amount of care has to be taken to get it of the right nsity, otherwise the subsequent counting may be different and even llacious.

Technique.—The technique employed was that of Wright, and as scribed by him in various publications.[1] The blood was collected a glass capsule with a recurved limb. When the blood coagulates the be is hung in a centrifuge, which expedites the separation of the serum.

2. The leucocytes were in all cases obtained from myself by pricking e finger and allowing the blood to run into a capsule containing sodium rate (1 per cent.) dissolved in ·85 per cent. sodium chloride. A blood-psule with the recurved limb bent at right angles to the plane of the dy of the capsule is very suitable for this purpose, as the citrate solution n be readily run into the capsule without any risk of contamination the solution. When the blood has entered the citrate solution the raight end of the capsule is sealed in a flame, and, when cool, the capsule hung in the centrifuge whereby the blood corpuscles, red and white, e carried to the bottom, sharply separated from the citrated plasma ove. When the deposition of the corpuscles is complete, the capsule cut across with a bone-pliers or file, the citrated plasma is removed by small pipette with a teat, and the thick deposit of corpuscles is then ansferred to a large 20 c.c. tube full of ·85 per cent. saline solution, in der to wash the corpuscles free of traces of plasma. The corpuscles e then brought down in the centrifuge, the supernatant liquid is pipetted f, and the corpuscles are ready for use.

3. An emulsion of tubercle bacilli is made by grinding in an agate ortar a small quantity of a bacillary mass with a solution of salt con-ining 1 : 1000 NaCl.

The preparation of this emulsion is a point of great importance, d great care is required in order to get it of the most suitable density. ie bacilli used were those obtained on the filter-paper in the manufac-re of Koch's tuberculin. After being sufficiently rubbed up in the ortar, the bacillary emulsion is put into a tube and centrifugalized, so to carry down the unloosened masses of bacilli, leaving above a milky nulsion of individual bacilli. If the emulsion is too thick, considerable fficulty may be experienced in subsequently counting the bacilli.

The serum, corpuscles, and bacillary emulsion having been prepared, series of capillary pipettes are taken in hand. A mark is made on the em of the capillary about ½ inch from the free end, and, by means of a

[1] *Roy. Soc. Proc.*, 1903, vol. lxii, p. 357 (pp. 76–78 *supra*).

teat, three volumes of serum, one volume of bacillary emulsion, an three volumes of blood-corpuscles are aspirated into the stem of th pipette. This mixture should then be blown out and aspirated in, b means of the teat, several times to ensure proper mixing. When this ha taken place, the end of the capillary is sealed, the wide end marke with the name of the patient or the number, and the time noted. Th pipette, with its contents, is then placed in the incubator at 37° C. anc after fifteen minutes, it is removed and the contents transferred to slide on which the subsequent count is to be made. The slide is prepare by roughening its surface by fine emery-paper (Hubert, No. 00) as firs suggested by Wright, this method invariably giving, with the cheapes slides, excellent films without any other preparation in the way c cleaning, boiling, etc.

When the mixture of bacilli, corpuscles and serum is placed on on end of the slide the actual film is made by spreading it out by means of second slide. This is best done in such a way that the second slide applied about three-quarters across the first, so that a free edge is obtaine to the film. In this free edge can be obtained a large number of leucocyte and it is the only part of the film which it is necessary to examine. Th film should not be made too thin.

The film is fixed with perchloride of mercury (sat. sol.), and is the stained. Perhaps the best results are obtained by staining, first of al with Ehrlich's haematoxylin, washing in water, hot carbol fuchsin, per cent. anilin chlorhydrate, alcohol, water. The film is then drie with blotting-paper, and is ready for the determination of the ingeste bacilli.

Staining with hot carbol fuchsin, subsequent decolourization in per cent, H_2SO_4, and counterstaining with Loeffler's blue, may also b used, but seems to be more uncertain in its results.

The free edge of the film is then placed under the microscope and i very polynuclear leucocyte encountered the number of bacilli is countec noted on a sheet of paper, and, after some thirty-five to fifty leucocyte have been examined, the average number of bacilli is struck, and th results compared when different sera are tested.

Determination of the Opsonic Content of Normal Individuals.

To determine the question of a variation in the opsonic index c healthy people I have examined sixty-six normal sera. Thirty-fou were obtained from robust medical students at ages of 18 to 30 years The remaining thirty-two were from nurses at the London Hospital, all c whom were presumably healthy. Up to the present we have no means c constituting the necessary standard of normality except by taking th serum of an individual presumably normal, i.e. an individual of soun constitution, devoid of tubercular infection and without inherited dis position to the disease. The opsonic index, it will be remembered, i

tained by dividing the number of tubercle bacilli taken up per leucocyte the presence of any serum by the number of bacilli taken up per leucyte in the presence of the serum of a normal individual, which latter regarded as unity. Thus—

B. taken up per leucocyte in the presence of a given serum=4
" " " " " " " normal=8

$$\frac{4 \text{ T. B.}}{8 \text{ T. B.}} = \text{opsonic index } \cdot 5.$$

e reason why the opsonic *index* is taken is that in different experiments the emulsions of bacilli vary in density, it would be useless to give erely the *number* of bacilli engulfed. One and the same serum, tested two different emulsions, will yield entirely different records.

Opsonic index of thirty-four medical students compared with the rum of the writer=1·0.

1	case	= 1·2
13	"	= 1·0
6	"	= 0·98
3	"	= 0·97
2	"	= 0·96
6	"	= 0·90
2	"	= 0·85
1	"	= 0·80

34 cases. Average opsonic index = 0·965.

Opsonic index of thirty-two healthy hospital nurses :—

5	cases	= 1·1
13	"	= 1·0
1	"	= 0·98
2	"	= 0·97
1	"	= 0·95
6	"	= 0·90
1	"	= 0·85
3	"	= 0·80

32 cases. Average opsonic index = ·969.

Taking the two series together, we get an average index of ·95 for the xty-six people, the variations in health ranging from ·8 to 1·2.

This corresponds closely to the results obtained by R. H. Urwick a thesis presented for the degree of M.D. Cambridge. This work, arried out at St. Mary's Hospital, showed—

8	cases	= 1
3	"	= ·94
3	"	= ·90
1	"	= ·8
2	"	= 1·1
3	"	= 1·2

20 cases. Average opsonic index = 1·006.

Taking the two series together, we have eighty-six healthy peopl with an index of 0·97. It may be assumed, therefore, that the sera o normal individuals, males or females, are almost identical, an index belov ·8 being rare or pathological.

Determinations of the Opsonic Index of Individuals Suffering from Lupus.

Turning to the question of the opsonic index of individuals sufferin from lupus, I have examined, as stated above, 150 cases. In thi series were cases of the mildest character up to the most severe forms which had lasted and defied treatment for as long as forty years. Com pared with the average opsonic index of ·97 obtained from normal people the average for the 150 cases of lupus is ·75, the cases being distribute as follows :—

Opsonic Index.	Number of Cases.	Percentage.
Between ·2–·3	3	2 per cent.
„ ·3–·4	3	2 „
„ ·4–·5	21	14 „
„ ·5–·6	29	19·6 „
„ ·6–·7	33	22 „
„ ·7–·8	22	14·8 „
„ ·8–·9	18	12 „
„ ·9–·10	7	4·6 „
„ ·1–·14	14	9·3 „

75 per cent. of the cases are below the lowest range of normal limit, viz. ·8

As was stated above, these patients suffering from lupus have attende the Skin Department of the London Hospital, and especially for th purpose of X-ray or Finsen light treatment. Data are available whereb we may compare the results obtained by these therapeutic measures wit the determinations of the opsonic index. As many of the cases are sti under treatment, I have tabulated only those in reference to whom a opinion may be formed. This opinion has been given by Dr. Sequeira who has had, in connexion with the Finsen therapy, an unusually larg experience of lupus. In the following tables the cases are divided int two series, viz., those with an opsonic index below ·7 and those with a index above ·9.

Cases of Lupus with an Opsonic Index below ·7.

Patient.	Opsonic Index.	Remarks.
R. Q.— (6) .	·44	Lupus vulgaris of nose ; treatment scraping, X rays result ?
J. B.— (20) .	·40	Lupus vulgaris ; father died of phthisis ; Finsen therap 4 years ; slow but great improvement.

Patient.	Opsonic Index.	Remarks.
W.— (13).	·5	Lupus 4 years, spreading in spite of X-rays for 1 year.
K.— (14)	·3	Lupus 11 years; Finsen therapy 3 years; not much improved.
B.— (10).	·55	Lupus; great tendency to relapse after Finsen treatment.
H.— (21).	·43	Lupus not severe; healed January, 1905.
H.— (16).	·54	Lupus 14 years; Finsen therapy 3 years; "can just be kept under and prevented from spreading."
C.— (18).	·5	Mother and aunt died of phthisis; lupus on right cheek 18 years; scraped 3 times; Finsen light 2 years; seems healed (December 21, 1904).
P.— (16)	·46	Lupus of face; cured with Finsen light.
A.— (22).	·55	No phthisis in family; lupus for 6 years; Finsen light for 1 year; doing well.
N.— (35).	·47	No phthisis in family; lupus 7 years; Finsen light 1 year; doing well.
[.—(27) .	·53	Phthisical family history; lupus 10 years; Finsen therapy; result, cured; relapse.
P.— (17) .	·25	Lupus 6 years; doing well under light treatment.
S.— (22).	·45	Father and paternal uncles and 1 sister all died from phthisis; lupus 22 years; scraped 12 times; elbow excised; Finsen light 8 months; doing well.
B.— (13).	·3	Lupus very extensive on face and nose; X rays since 1900; Finsen light; little improvement.
B.—(41) .	·48	Lupus 16 years; scraped; Koch's tuberculin; apparently healed; relapsed as bad as ever in 6 months; Finsen light 1 year; cure; relapse in 2 months.
G.— (49).	·46	Two children died from phthisis; lupus 11 years; scraped; X rays; almost healed; recurrence; again under treatment.
M.— (37).	·55	Phthisical family history; lupus for 14 years, very extensive; scraped 16 times; Finsen light not very successful.
R. . .	·60	No phthisis in family; lupus 8 years, very extensive; scraped several times; Finsen therapy for 2 years; nodules still remaining.
W.— (25)	·60	No phthisical history; lupus both cheeks; cured by Finsen light in 6 months; recurrence.
R.— (17).	·65	Lupus 13 years; Finsen light 3 years; improvement very slow.
F.— (20).	·60	Father died of phthisis; sister has phthisis; lupus of face, mouth, larynx, right arm for thirteen years; some improvement under X-rays and Finsen light.
B. . .	·68	Marked phthisical history; lupus 34 years; Finsen light for 3 years; slow improvement.
C.— (26).	·62	Lupus 22 years; Finsen therapy 2 years; slow improvement.
S.— (16).	·66	Phthisical history; lupus 15 years; Finsen light on and off for 5 years; some nodules still present (1905).
C.— (48).	·65	Sister had phthisis; lupus 9 years on nose and face; Finsen light for 4 years; a few small spots of lupus still remain (1905).
W.— (19)	·6	No history of phthisis; lupus 10 years on nose, palate, gums, cheeks; treated with lactic and trichloracetic acids; Finsen light since 1901; iodides; much better.
B.— (33).	·6	No phthisis in family; lupus 19 years; scraped 19 times; cauterised about 100 times; 600 sittings Finsen light; great improvement.

Cases with an Opsonic Index above 9.

Patient.	Opsonic Index.	Remarks.
M. S.—(19) .	·9	Phthisis on paternal side ; lupus 18 years ; Finsen light cured.
L. C.—(29) .	1·1	Lupus began in scar of ulcer from tubercular gland at ag of 14 ; in 1905 very large lupus patch over jaw ; Finse light 6 months ; nearly healed.
F. S.—(33) .	·9	Phthisis in family ; patient had hip disease at 9 ; lupu for 8 years ; nose severely affected ; Finsen light an X rays ; face excellent.
J. S.—(16) .	·9	No phthisis ; lupus 15 years ; Finsen light ; cured.
B. W.—(42).	·9	No phthisical history ; lupus 40 years ; marked improve ment under Finsen light.
M. T. . .	·9	Lupus extensive on face ; Finsen light ; cured ; no re currence.
M. B.—(28) .	1·1	Lupus 24 years ; left cheek entirely involved ; Finse light for 2 years ; quite healed ; scar excellent.
S. M.—(19) .	·89	Lupus 12 years ; very severe case ; much improved unde Finsen light.
A. S. . .	1·1	Lupus 13 years ; " much improvement."
T. S. . .	1·1	Lupus 11 years ; X rays ; great improvement.
T. A.—(27) .	1·4	Lupus 22 years ; very severe case ; almost cured.
R. S.—(25) .	·92	Lupus of face and stump of thigh ; " has done well."
A. J.—(12) .	1·4	Five uncles died of phthisis ; lupus 11 years ; scrape and cauterised several times ; Finsen light ; resul almost cured.
M. H.—(16).	1·0	Father and two sisters died of phthisis ; very extensiv lupus ; heals very slowly under Finsen therapy.
F. H.—(16) .	1·0	Lupus 10 years, nose, left foot, thigh ; not improving.
A. J.—(25) .	1·0	Lupus 17 years ; Finsen light 1 year ; great improvement

An examination of these tables shows, with few exceptions, tha where the opsonic index is well below the normal limit, the most approve methods of treatment by X rays or Finsen light have little power t stamp out the disease, whereas with indices in the normal limit or abov it the clinical impression is that the cases do well. The method by whic the exposure to light in Finsen's treatment leads to beneficial results ha been the subject of much inquiry. The bactericidal action of ligh known since the researches of Downes and Blunt, Buchner, and others has been assumed to be the explanation, but in the hands of Finsen' collaborators it has been found that this action, even *in vitro*, is a ver superficial one. When the rays have to penetrate the skin into the livin tissues the state of affairs is very different, as a large amount of the ligh is absorbed. As a result of experiments, both in man and in animals Klingmüller and Halberstaedter have recently shown that the tubercl bacillus in the skin is not killed after seventy minutes' exposure to th light. Lesser and others have assumed that the healing properties o Finsen light are due to the reaction set up in the tissues by the rays. Th results obtained above go to support the view that ultra-violet rays fron a Finsen lamp do not exert a potent effect on the tubercle bacillus itsel

ıerwise it would not matter what the opsonic content of the serum ..s, so long as the bacilli were killed at any given spot exposed to the ht. It seems not at all improbable that, in addition to the tissue ıction, an important rôle in the cure of lupus by Finsen light is played · the blood " determination "—i.e. the congestion and exudation which cur after exposure. If the plasma is deficient in opsonin, the result ›on the tubercle bacillus would manifestly be less than where a large ıantity of opsonin was present. This would also suggest that consider-le benefit might be acquired even in the intractable cases by raising the sonic index and then exposing the infected areas to light.

On the Possibility of Determining the Presenc or Absence of Tubercular Infection by th Examination of a Patient's Blood an Tissue Fluids.[1]

By A. E. Wright and Staff-Surgeon S. T. Reid, R.N.

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Classification of Tubercular Cases into strictly Localized Cases, and Cases which a associated with Constitutional Disturbance—Data with regard to the Tubercu opsonic Power in Cases of Strictly Localized Tuberculosis—Data with regard the Tuberculo-opsonic Power of the Blood in Cases of Tuberculosis associat with Constitutional Disturbance—Suggested Interpretation of the Differe Findings in these two Categories of Cases—Exploitation of the Data summariz above as an Aid in the Diagnosis of Tubercular Infection—Discrimination Tubercular Blood from Normal Blood by the aid of the Phagocytic Test conduct with Serum which has been subjected to a Temperature of 60° C.—On two oth Methods by which a Diagnosis of Tubercular Infection can be arrived at excluded—Diagnosis of Tubercular Infection by the Aid of Measurements of t Opsonic Power carried out in Connexion with the Inoculation of Tuberculin f Diagnostic Purposes—Diagnosis of Tubercular Infection by the Comparison the Opsonic Power of the Patient's Blood with the Tuberculo-opsonic Pow of the Fluids Derived from the Focus of Infection.

In the present communication we propose (*a*) to set forth certain co clusions arrived at after the study of the tuberculo-opsonic power of t blood in a very considerable number of tubercular patients; (*b*) to sho that we have in the measurement of the tuberculo-opsonic power of t blood and tissue fluids a method which may be exploited in the diagno of tubercular infection.

Technique Employed.—The technique employed by us in the measu ment of the tuberculo-opsonic power of the blood was essentially th described by one of us in conjunction with Douglas.[2] In each case t white corpuscles required for the tests were derived from blood from t finger received into a solution of 0·5 per cent. citrate of soda in 0· NaCl, and rewashed after centrifugalization in a considerable volume 0·85 NaCl, and then again centrifugalized. Of the "blood cream

[1] Reprinted from the *Proceedings of the Royal Society*, B, vol. lxxvii, 1906.

[2] *Roy. Soc. Proc.*, vol. lxxii. (pp. 76–78 *supra*).

›tained by skimming off the upper layer of the corpuscular sediment, ıe portion was in each case mixed in a capillary tube with one volume ; serum and one volume of a suspension of tubercle bacilli which had ›een centrifugalized in such a manner as to free it from bacillary clumps. fter incubation at 37° C. for fifteen to twenty minutes films were made on ides prepared with emery paper.[1] These films were, after fixture . saturated corrosive sublimate, stained with boiling carbol-fuchsin, ›colourized with 2 per cent. sulphuric acid, and counter-stained with ethylene blue after washing in 1 in 1000 sodium carbonate. The standard ; comparison employed was obtained by mixing in each case the same blood cream" and tubercle suspension with "pooled normal serum."[2] his "pooled serum" was obtained by mixing equal volumes of the sera ; six to eight healthy students or laboratory workers. We have found ıat the opsonic power of such a "pooled serum" corresponds to the rithmetical mean of the opsonic indices of its component sera.

lassification of Tubercular Cases into Strictly Localized Cases, and Cases which are associated with Constitutional Disturbance.

Cases of tubercular infection distribute themselves in a natural ıanner under two headings. Into one category would fall the patients ho are the subjects of a strictly localized infection unaccompanied by ıything in the nature of constitutional disturbance. Cases where the ıfection is limited to one or more lymphatic glands ; further, most cases ; lupus, most cases of tubercular abscess in the subcutaneous tissue, ıbercular affections of the joints, and, lastly, many stationary or only owly progressing cases of tubercular phthisis, fall into this category.

Into another category would fall patients who are suffering from ıore generalized tubercular infections associated with constitutional sturbance. This group consists in large part of cases of pyrexial pul-ıonary tuberculosis. With these may be classed certain other cases of ‹tensive or widely disseminated tuberculosis.

ata with regard to the Tuberculo-opsonic Power in Cases of Strictly Localized Tuberculosis.

The opsonic index is here low and uniformly low—in exceptional cases

[1] Wright, *Lancet*, July 9, 1904.

[2] While in this research pooled serum was employed, in order to provide against ıy chance variation of our bloods under the physical strain entailed by the work, is to be noted that the observations of Urwick, conducted in this laboratory, *ritish Medical Journal*, July 22, 1905, and the more extensive series of investi-ıtions carried out by Bulloch at the London Hospital (*Medico-Chirurg. Soc. Proc.*, 905), and Lawson and Stewart at the Banchory Sanatorium (loc. cit.), have ›nclusively shown—(*a*) That the tuberculo-opsonic power of the blood does not . health range below 0·9 or above 1·1 ; and (*b*) that the bloods of A. E. W., . R. D., and others which have hitherto in this laboratory furnished a standard ; comparison are, from the point of view of their tuberculo-opsonic power, ›pically normal bloods.

as low as one-sixth of the normal. Our findings in a series of cases ▮ stricty localized tubercular infection are appended in tabular form belo▮

Table I.—*Showing the Tuberculo-opsonic Index in a Series of Cases ▮ strictly Localized Tuberculosis.*

Serial No.	Initials, or as the case may be, Initial of Patient.	Nature and Seat of the Infection.	Tuberculo-opsonic Index.
1	J. R.	Tubercle of testis	0·65
2	A.	Caries, lower end of femur	0·7
3	C. S.	Tubercular ulceration, dorsum of hand	0·86
4	A. B.	„ iritis	0·51
5	B. C.	„ glands	0·4
6	E. M.	„ ulceration of legs. 13 years' duration	0·17
7	H. W.	„ glands (neck)	0·82
8	D. W.	„ „ „	0·64
9	S.	„ ulceration of legs	0·49
10	L. B.	„ glands (abdominal)	0·13
11	B.	„ kidney	0·75
12	M. H.	„ cystitis	0·85
13	D. B.	„ glands. Extirpated and reappeared	0·85
14	M. O. E.	„ glands	0·6
15	W.	Psoas abscess	0·75
16	C. H.	„ „	0·65
17	W.	Tubercular glands (neck). 18 months' duration	0·47
18	C.	„ wrist	0·85
19	U. R.	„ glands (neck)	0·7
20	A. H.	„ glands. Extirpated and reappeared	0·54
21	P.	„ peritonitis	0·6
22	R.	„ abscesses and glands	0·6
23	E.	Lupus	0·6
24	R.	Tubercle of testes and bladder	0·72
25	P.	Tubercular peritonitis	0·7
26	H.	„ caries of fibula	0·6
27	C.	Tubercle of kidney	0·88
28	W.	Tubercular disease of knee	0·6
29	T.	„ glands (neck) recurrence	0·66
30	S.	„ ulcer of foot	0·49
31	C.	„ disease of knee	0·75

Data with regard to the Tuberculo-opsonic Power of the Blood in Case▮ of Tuberculosis associated with Constitutional Disturbance.

In the cases here in question the opsonic index of the blood is con▮ tinually varying. The range of its fluctuation is from considerabl▮ under the normal to twice, or more, the normal height.

Striking examples of the variation of the opsonic index in connexio▮ with acute tubercular phthisis are furnished in the paper of our fellow▮ worker, R. H. Urwick, already referred to.

[1] Loc cit.

The following are instances of similar variation occurring in the ›jects of other forms of tubercular infection :—

ample 1.—Child with Tubercular Caries of the Fibula, associated with Constitutional Disturbance.

Dates of Blood Examinations.	Tuberculo-opsonic Index.
11.9.05	1·45
14.9.05	1·71
19.9.05	1·3
28.9.05	0·98
30.9.05	Operation, fibula scraped.
2.10.05	1·73[1]
3.10.05	1·13
10.10.05	1·3

ample 2.—Adult Patient with Tubercular Caries of the Spine and Constitutional Disturbance.

Date of Blood Examination.	Tuberculo-opsonic Index.	Remarks.
19.5.05	0·65	Temperature disturbance and pain associated with development of abscess.
20.5.05	1·4	
22.5.05	1·3	
23.5.05	1·0	Temperature returns to normal in association with spontaneous discharge of abscess.
24.5.05	0·8	

ample 3.—Adult Patient with extensive Psoas Abscess and Generalization of Tubercle. Case has since terminated fatally.

Dates of Blood Examinations.	Tuberculo-opsonic Index.
8.2.05	2
9.2.05	2·4
11.2.05	0·6

ggested Interpretation of the Different Findings in these two Categories of Cases.

The explanation of the difference in the condition of the blood in se two contrasted categories of cases is probably the following : The ıdition of low opsonic power which is associated with strictly localized erculosis is almost certainly a condition which has preceded and has nished the opportunity for infection. The fact that the opsonic ex continues persistently low after infection has supervened, while can invariably be raised by appropriate inoculation,[2] indicates that

[1] A rise in the opsonic power similar to this here registered has been repeatedly erved by us in connexion with the stirring up by surgical interference of tubercular .

[2] Exactly the same statements hold true with regard to the staphylo-opsonic ver in localized staphylococcus infections (furunculosis, sycosis, etc.).

the machinery of immunisation with which the organism is furnis is not, under the conditions which obtain in strictly localized tubercu infections, spontaneously called into play.

The constant fluctuation in the opsonic power of the blood in ca of active pulmonary tuberculosis and other active forms of tubercu infection furnishes—as we can hardly doubt—evidence of a perio conveyance of tubercular elements into the blood; and of a respo to such stimulation on the part of the machinery for immunisation. T low opsonic indices registered in connexion with active tubercul would, in other words, be "negative phases" such as supervene—as of us has shown—upon the inoculation of all vaccines; the high opso indices would be "positive phases," such as normally succeed upon negative phases just mentioned; and the normal opsonic indices wo correspond to periods of transition between negative and positive pha or, as the case may be, to periods in which the blood is returning afte positive phase to the condition *quo ante*.

The life of a patient with any really active form of tuberculosis wou in conformity with this view, be a life of alternating negative and posit phases: the favourable or unfavourable event of the infection being each case determined by the adjustment or want of adjustment of auto-inoculations (with respect to dosage and interspacing) with particular patient's capacity for immunising response.

Having now to a certain extent cleared the ground, we may pass to consider the question of the diagnosis of tubercular infection by me of the measurement of the opsonic power of the blood.

Exploitation of the Data summarized above as an Aid in the Diagn of Tubercular Infection.

Consideration will make clear that the data obtained by the measu ment of the opsonic power in cases of doubtful diagnosis may, w adjudicated upon in the light of the data obtained in connexion w undoubted cases of tuberculosis as given above, furnish material admitting or rejecting the diagnosis of tubercular infection. We n formulate in connexion with this matter the following propositions

(1) Conclusions which can be arrived at when we have at disposal results of a series of measurements.

(a) *Where a series of measurements of the opsonic power of the bl reveals a persistently low opsonic power with respect to the tubercle bacil it may be inferred, in the case where there is evidence of a localized bacte infection which suggests tuberculosis, that the infection in question is tu cular in character.*

(b) *Where repeated examination reveals a persistently normal opso power with respect to the tubercle bacillus, the diagnosis of tubercle may i probability be excluded.*

Illustrative case: A. B.—Case diagnosed as tubercular cystitis on

idence of pus in the urine, of the cystoscopic appearances and general sturbance of health. The measurement of the tuberculo-opsonic power the blood yielded the following results :—

Date of Blood Examination.	Tuberculo-opsonic Index.
2.3.05	0·98
14.4.05	0·99
28.4.05	1
18.5.05	1
19.5.05	1·1
2.10.05	0·97

The inference that the cystitis and disturbance of health was not of bercular origin was confirmed (*a*) by the fact that an extensive series bacteriological examinations prolonged over many months revealed every case the presence of proteus in large numbers, while the tubercle cillus was never found, even when examined for by the inoscopic thod of Jousset ; (*b*) by the fact that the patient's blood possessed, terior to treatment with regard to the proteus, an agglutinating power ich was three times higher than the normal ; and (*c*) by the fact that y striking amelioration of the cystitis, and a complete return to health s been obtained as the result of the inoculation of a proteus vaccine.

(c) *Where there is revealed by a series of blood examinations a constantly ctuating opsonic index the presence of active tuberculosis may be inferred.*

C. D.—A case of severe chronic urticaria of unknown aetiology. The asurement of the tuberculo-opsonic power of the patient's blood yielded following results :—

Date of Blood Examination.	Tuberculo-opsonic Index.
20.5.05	1·3
26.5.05	1·3
16.6.05	0·86
20.6.05	1·27

The inference drawn from these data that the patient was suffering m some active form of tuberculosis was confirmed (*a*) by the discovery an independent observer of a lesion in the apex of one lung ; (*b*) by subsequent development of an abscess of an obviously tubercular racter ; and (*c*) by the marked improvement in health which has owed upon inoculation with tubercle vaccine.

(2) Conclusions which may be arrived at where we have at disposal result of an isolated blood examination.

(a) *Where an isolated blood examination reveals that the tuberculo-onic power of the blood is low, we may—according as we have evidence a localized bacterial infection or of constitutional disturbance—infer h probability that we are dealing with tuberculosis—in the former case h a localized tubercular infection, in the latter with an active systemic ection.*

(b) *Where an isolated blood examination reveals that the tuberculo-onic power of the blood is high, we may infer that we have to deal with a temic tuberculous infection which is active, or has recently been active.*

(c) *Where the tuberculo-opsonic power is found normal, or nea normal, while there are symptoms which suggest tuberculosis, we are warranted, apart from the further test described below, in arriving at a po tive or a negative diagnosis.*

Discrimination of Tubercular Blood from Normal Blood by the aid of Phagocytic Test conducted with Serum which has been subjected a Temperature of 60° C.

The further criterion to which reference was made in the preced paragraph is the following :—

When a serum is found to retain in any considerable measure, afte has been heated to 60° *for ten minutes, its power of inciting phagocytosis, may conclude that " incitor elements "*[1] *have been elaborated in the organis either in response to auto-inoculations occurring spontaneously in the cou of tubercular infection, or, as the case may be, under the artificial stimu supplied by the inoculation of tubercle vaccine.*

A typical selection from the very extensive body of observations wh furnishes the basis of the above statement is presented in Tables II a III.

Table II.—*Showing that the Normal Serum, after it has been exposed t Temperature of* 60° C. for *ten Minutes no longer incites Phagocytosis*

Serial Number of the Observation.	Derivation of the Serum.	Unheated Serum.		Heated Serum.	
		Phagocytic count. (Number of Bacteria ingested divided by Number of Leucocytes examined.)	Tuberculo-opsonic Index.	Phagocytic Count. (Number of Bacteria ingested divided by Number of Leucocytes examined.)	Tubercul opsonic Index.
1	Healthy man .	(104/40) = 2·6	Taken as 1	(13/40) = 0·32	0·125
2	" " .	(96/40) = 2·4	" 1	(8/40) = 0·2	0·08
3	Pooled serum of six healthy men	(247/36) = 6·8	" 1	(30/50) = 0·6	0·09
4	Healthy boy. .	(250/39) = 6·4	" 1	(15/40) = 0·4	0·06
5	" " . .	(214/30) = 7·0	" 1	(10/40) = 0·47	0·06
6	Pooled serum of eight normal men	(60/50) = 1·2	" 1	(2/20) = 0·1	0·08
7	Healthy man .	(55/40) = 1·4	" 1	(0/40) = 0·0	0·00
8	Pooled serum of six healthy men	(132/30) = 4·4	" 1	(3/30) = 0·1	0·1

[1] The term " incitor elements " (Latin, *incito,* I urge forward, I hasten, I bri into rapid movement) is here employed in lieu of a more specific term, in or not to prejudge the mode of action of the element in the heated serum which p motes phagocytosis. The nature of the incitor element is considered in the ne following communication (pp. *infra*).

[2] In order to avoid the fallacies associated with spontaneous phagocytosis (*v* the next following communication) the observations which are recorded in t and in the subsequent table were in each case made by mixing the volume of serum with one volume of corpuscles, washed in 0·85 per cent. NaCl solution a one volume of tubercle bacilli suspended in a 1·5 per cent. NaCl solution. In t manner a salt content of over 1 per cent. NaCl was achieved in the phagocy mixture.

BLE III.—*Showing that an element which incites Phagocytosis is contained in the Heated Serum of Patients who are the Subjects of an Active Systemic Tubercular Infection, or who have been subjected to Inoculations of a Tubercle Vaccine. The Sera, like those which are in question in Table* II, *were in each case heated to* 60°*C. for ten Minutes.*

ial nber f ser- ion.	Nature of Infection.	Unheated Serum.		Heated Serum.	
		Phagocytic Count. (Number of Bacteria ingested divided by Number of Leucocytes examined.)	Tuberculo-opsonic Index (determined by Comparison of Phagocytic Count with that obtained with Pooled Blood of Healthy Men.)	Phagocytic Count. (Number of Bacteria ingested divided by Number of Leucocytes examined.)	Tuberculo-opsonic Index (determined by Comparison of Phagocytic Count with that obtained with Unheated Pooled Blood of Normal Men).
1	Tubercular caries of hip	—	1·5	—	0·4
2	Tubercul'r phthisis	(125/20)=6·2	1·4	(113/30)=3·7	0·8
3	,, ,,	(152/30)=5·0	1·2	(96/30)=3·2	0·72
4	,, ,,	(98/30)=3·2	1·0	(20/65)=0·3	0·1
5	Tubercular peritonitis	(144/30)=4·8	1·4	(103/30)=3·4	1·0
6	Tubercular peritonitis	(142/30)=4·7	1·4	(16/50)=0·3	0·09
7	Phthisis and tubercular glands	(113/40)=2·8	1·1	(79/50)=1·6	0·6
8	Tubercular caries of hip	(110/30)=3·6	1·0	(85/30)=2·8	0·8
9	Tubercular abscess of kidney	—	1·7	(26/30)=0·8	0·4
0	Lupus under treatment by inoculation of tubercle vaccine	(34/10)=3·4	0·7	(49/30)=1·6	0·33
1	Tubercular ulcer of leg under treatment by inoculation of tubercle vaccine	(249/40)=6·2	1·2	(149/40)=3·7	0·7
2	Tubercle of kidney under treatment by inoculation of tubercle vaccine	(68/40)=1·7	1·5	(77/40)=1·9	1·7
3	Tubercular glands and abscess under treatment by inoculation of tubercle vaccine	(59/40)=1·5	1·4	(36/40)=0·9	0·8
4	Tubercular cystitis under treatment by inoculation of tubercle vaccine	(97/50)=2·0	—	(43/30)=1·4	—
15	Phthisis . . .	—	—	(26/30)=0·8	—
16	,, . . .	—	—	(9/5)=1·8	—

It will be seen from these tables that in practically every case wl a reaction to tubercular infection may be assumed to have taken pl evidence of that reaction can be obtained by conducting the phagoc test with serum which has been heated to 60° C. for ten minutes.

The observations numbered 15 and 16 respectively have it may noted, been introduced into the table with the special design of show the very simple nature of the investigation which is required for diagnosis of tubercle in the case where that infection has called fort reaction of immunisation.

The following observations, which we owe to our fellow-worker G. W. Ross, bring out in an instructive manner the trustworthiness the phagocytic test with heated serum as applied in this its simp form :—

Case 1.—*Girl, æt. Six Years, tentatively diagnosed Pulmonary Phthis*

Phagocytosis obtained with the serum, heated for ten minutes 60° C. and employed in a phagocytic mixture containing over 1 per ce NaCl.

The verdict of tubercular infection of the lung which was based this was confirmed on *post-mortem* examination.

Case 2.—*Man, æt.* 41, *Tentative Diagnosis, Pleurisy due to Maligna Disease, or Tubercular Pleurisy.*

No phagocytosis obtained with the serum, heated for ten minutes 60° C. and employed in a phagocytic mixture containing over 1 per ce NaCl.

The verdict of pleurisy due to malignant disease, which was based this, was confirmed on *post-mortem* examination.

Case 3.—*Case Tentatively Diagnosed Miliary Tuberculosis or Maligna Endocarditis.*

No phagocytosis obtained with the serum, heated for ten minu to 60° C. and employed in a phagocytic mixture containing over 1 p cent. NaCl.

The verdict of malignant endocarditis which was based on this w confirmed on *post-mortem* examination.

Observation 4.—*Case Diagnosed Miliary Tuberculosis.*

No phagocytosis obtained with the serum, heated for ten minut to 0° C. and employed in a phagocytic mixture containing over 1 p cent. of NaCl.

The *post-mortem* examination revealed a complete absence of tuber-
ılar lesions and a healing typhoid ulcer[1] in the ileum.

n two other Methods by which a Diagnosis of Tubercular Infection can be arrived at or excluded.

In addition to the methods which have been already considered, there ·e two further methods which can be exploited in connexion with the agnosis of tubercular infection. The first of these is applicable where e desire to supplement the often ambiguous data furnished by the clinical ·mptoms in the case of inoculations of tuberculin undertaken for diag- ɔstic purposes. The second is applicable where we can obtain, in addition the patient's blood, also lymph, or, as the case may be, pus from the at of infection.

iagnosis of Tubercular Infection by the Aid of Measurements of the Opsonic Power carried out in Connexion with the Inoculation of Tuberculin for Diagnostic Purposes.

Already, three years ago,[2] in connexion with a paper on staphylococcus oculations as applied to the treatment of acne, furunculosis, and sycosis, tention was directed by one of us to the close analogy between the berculin reaction of Koch and the local inflammation and general nstitutional disturbance which may supervene when a patient whose ssues are extensively invaded by the staphylococcus is inoculated with e corresponding vaccine in such a manner as to develop a pronounced gative phase.

The association of a negative phase with a reaction similar to that nveniently spoken of as the *tuberculin reaction*, suggested to us the opriety of inquiring whether the true tuberculin reaction, as seen after e injection of Koch's old tuberculin into a tubercular patient, was also sociated with a negative phase.

The opportunities for investigating the question which have presented emselves have not yet been sufficiently numerous to allow of our rmulating a final answer to this question. The observations which are t forth below seem to us to suggest that the development of a negative ıase, with a dose of tuberculin smaller than that which would produce is result in a healthy patient, may prove to be an index of tubercular fection. Such a conclusion would be in harmony with our experience connexion with the therapeutic inoculation of tubercle vaccine (new berculin). We find in this connexion that the negative phase super- nes upon very much smaller doses and persists much longer in the case ıere the patient is the subject of extensive infection than in the contrary ses.

[1] A negative agglutination reaction had been obtained in this case.
[2] *Lancet*, March 29, 1902 (pp. *infra*).

Observation 1.—Case diagnosed, Tubercular choroiditis.

Date.	Tuberculo-opsonic Index.	Clinical Data.
26.4.05	0.9	—
5 milligrammes old tuberculin inoculated.		
29.4.05	0·29	Some constitutional r
28.4.05	0·95	action, *t.* 100° F.

Observation 2.—Case diagnosed, Lupus erythematosus.

Date.	Tuberculo-opsonic Index.	Clinical Data.
12.1.05	0·73	—
Inoculation of 1 milligramme old tuberculin.		
13.1.05	0·85	No rise of temperature
17.1.05	1·6	constitutional or local
26.1.05	0·5	action.

Observation 3.—Case diagnosed, Lupus erythematosus.

Date.	Tuberculo-opsonic Index.	Clinical Data.
10.4.05	0·66	—
Inoculation of 5 milligrammes of old tuberculin.		
11.4.05	0·7	Quite insignificant cons
12.4.05	1·2	tional disturbance.
14.4.05	0·85	

Observation 4.—Case diagnosed as Lupus vulgaris.

Date.	Tuberculo-opsonic Index.	Clinical Data.
10.4.05	0·55	—
Inoculation of 5 milligrammes of old tuberculin.		
11.4.05	1·1	Quite insignificant cons
12.4.05	1·0	tional reaction.
14.4.05	1·0	

Observation 5.—*Lupus Patient had been treated for many months by Therapeutic Inoculations of Tubercle Vaccine.*

Date.	Tuberculo-opsonic Index.	Clinical Data.
24.1.05	1·4	—
oculation of 30 milligrammes of old tuberculin.		
25.1.05	0·34	Severe constitutional and local reaction, *t.* of 103° F.
26.1.05	2·1	
27.1.05	1·7	

agnosis of Tubercular Infection by the Comparison of the Opsonic Power of the Patient's Blood with the Tuberculo-opsonic Power of the Fluids Derived from the Focus of Infection.

Attention has already been drawn by one of us, both in a research ıdertaken in conjunction with Lamb [1] and in a research undertaken in njunction with Douglas,[2] to the fact that we have in the actual focus of fection a lowered "bacteriotropic pressure" which accounts for the ltivation of the pathogenetic microbe in the interior of an organism ıich has at disposal in the circulating blood a considerable reserve of ti-bacterial substances. We propose here, in conclusion, to furnish rther illustration of the general law as enunciated above, culling our amples not alone from the observations we have made in connexion th tubercular infection, but also from observations made in connexion th other bacterial infections.

Observation 1.—*Case of Abscess in the Neighbourhood of the Appendix.* ood from the patient's finger and pus obtained from the abscess at e operation were examined, with a view to determining the nature of the ection.

	Phagocytic Counts.	
	With a Suspension of Tubercle Bacilli.	With a Suspension of Staphylococci.
um	2·3	4·5
ıid obtained from the pus by entrifugalization	0·1	1·9

The fact that the tuberculo-opsonic power of the patient's blood was e 23 times as great as that of the fluid obtained from the pus was ken as evidence that tuberculo-opsonic substances had been used up the pus and that the patient was suffering from a tubercular infec-

[1] *Lancet*, December 23, 1899 (pp. 36–44 *supra*).
[2] *Roy. Soc. Proc.*, vol. lxxiv, p. 157 (pp. 104–105 and 119–120 *supra*).

tion. It was inferred on similar grounds that he was also infec
by staphylococcus.

Observation 2.—*Case of Osteo-myelitis of the Femur.* Blood from
patient's finger and pus obtained from the abscess at the opera
were examined, with a view to determining the nature of the infectio

	Tuberculo-opsonic Index.	Staphylo-opsonic Ind
Serum	1·0	2·5
Fluid obtained from the pus by centrifugalization	1·1	0·9

The fact that the opsonic index of the patient's circulating blood
here normal to tubercle, while it was two and a half times greater t
normal with respect to the staphylococcus, was taken as evidence t
the patient was not infected with tubercle, and that he was infected
staphylococcus and had responded to that infection by a productior
immunising substances.

The fact that the tuberculo-opsonic index of the fluids obtained fr
the pus was the same as that of the blood, while the staphylo-opso
power was only two-fifths of that of the circulating blood, was taker
confirmatory evidence of the conclusion already arrived at. The f
that a copious culture of staphylococcus aureus was obtained from
pus, planted out with aseptic precautions at the operation, further c
firmed the diagnosis.

Observation 3.—*Case of Psoas Abscess.* Blood from the patie
finger and pus from the abscess were examined.

	Phagocytic Counts.	
	With a Suspension of Tubercle Bacilli.	With a Suspension o Staphylococci.
Serum	2·4	5·0
Fluid obtained from the pus by centrifugalization	1·23	1·2

The fact that the fluid obtained from the pus was impoverished
both tuberculo- and staphylo-opsonic substances as compared with
blood was taken as evidence of a combined infection by tubercle ba
and staphylococci. This inference was confirmed by the fact that
opsonic power of the blood with respect to both the micro-organisms l
in question was undergoing perpetual fluctuations.[1] The inference so
as it related to the staphylococcus was further confirmed by the f
that cultures of the micro-organism were obtained from the pus.

[1] For the variations registered in connexion with the tuberculo-opsonic po
vide supra, p. 153, where Example 3 refers to the patient here in question.

Observation 4.—*Case of Ascites with Grave Constitutional Disturbance in Man of* 30. Blood from the finger and ascitic fluid were examined on o occasions.

First Occasion.

	Tuberculo-opsonic Index.
Serum	1·05
Ascitic fluid	0·99

We reported upon this that the patient was not suffering from ercular peritonitis.

The clinical symptoms, the age of the patient, and the appearances seen at the operation appearing in contradiction with this verdict, l the ascites having reappeared, a second operation was performed, l a further sample of ascitic fluid was obtained for examination. At same time the clinical appearances were again observed, with the ult that there was now some wavering as to whether the original diasis could be upheld. The result of the phagocytic examination of the itic fluid, and of a sample of blood from the fingers were now as under :—

	Tuberculo-opsonic Index.
Serum	1
Ascitic fluid	1

In view of this result the verdict previously given was sustained.

A *post-mortem* examination, which followed in the course of a few eks, again threw doubt on the verdict, the naked-eye appearances being irely consistent with the theory of miliary tuberculosis affecting the itoneum and serous covering of the intestines. Microscopic examinan of the sections made through the miliary nodules revealed, however, ypical picture of miliary carcinoma. No primary carcinomatous focus l been discovered, though it was sought for, on *post-mortem* examination.

Observation 5.—*Case of Pleural Effusion.* Blood from the finger and d obtained by paracentesis of chest were examined :—

	Tuberculo-opsonic Index.
Serum	0·92
Pleural fluid	1·0

This was taken as evidence of the absence of tubercular infection.

Observation 6.—*Case diagnosed as Tubercular Peritonitis complicated h Pleurisy.* Blood from the finger was examined on two occasions. the second occasion, which was forty-eight hours after the first examtion, the patient's peritoneal and pleural fluids were also examined.

The results obtained by the phagocytic examination undertaken on second occasion were as follows :—

	Tuberculo-opsonic Index.
Serum	0·7
Peritoneal fluid	0·28
Pleural fluid	1

The results of the comparison of the peritoneal fluid with the se obtained from the blood withdrawn from the finger were taken as evid of tubercular infection of the peritoneum. Confirmatory evidence of tu cular infection was furnished further by the low tuberculo-opsonic p of the blood, and by the observed fluctuation in this index. Whe was measured two days previously, this index had worked out as 1

The fact that the opsonic power of the pleural fluid worked ou higher than the opsonic power of the serum was taken as evidence the pleural effusion had occurred at a period when the opsonic pow the blood was 1 or above 1.

The diagnosis of tubercular infection of the peritoneum and pl (and underlying lung) was confirmed at the *post-mortem* examinatio

Observation 7.—*Case of long-continued Suppuration of the Ant* associated with the presence in the pus of the pneumococcus and *Bacillus fusiformis* and *Spirillum buccae* of Vincent. The patient been treated by therapeutic inoculations of a pneumococcus vaccine. patient's serum and the antral pus were examined with a view to d mining whether the pneumococcus played any active part in conne with the continuance of the suppuration :—

	Pneumo-opsonic In
Serum	4·3
Fluid obtained from pus by centrifugalization . . .	0·3

The result was taken as evidence (*a*) that the pneumococcus pl an active rôle in connexion with the suppuration, and (*b*) that the prote substances which had been generated in the blood under the influen inoculation did not come satisfactorily into application upon the m organisms in the antrum.

Observation 8.—*Case of whitlow associated with the Formation Blister under the Nail.* Serum derived from the blood from a sound fi and blister fluid were examined.

	Staphylo-opsonic Index.
Serum	0·8
Blister fluid	0·3

The blister fluid yielded a pure culture of staphylococcus.

Observation 9.—*Rabbit in the Early Stages of Anthrax Infection.*—B obtained from the ear and lymph from the seat of inoculation examined.

	Anthraco-opsonic Index.[1]
Serum	1·7
Lymph	0·62

[1] Tested with a suspension of anthrax spores and compared with the s of a normal rabbit tested in the same manner.

It may be noted that all the difficulties and inaccuracies which are assoc with the employment of ordinary anthrax cultures in phagocytic experiment be satisfactorily evaded by the employment of suspensions of anthrax sp These, when stained with carbol fuchsin and decolourized by 0·25 per cent. sulp acid, represent absolutely ideal elements for enumeration.

Appendix.

urther Series of Observations showing that Phagocytosis is obtained with the Heated Serum of Patients who are the Subjects of a Systemic as distinguished from a strictly Localized Tubercular Infection, or who, being the subject of a strictly Localized Tubercular Infection, have been subjected to Inoculations with Tubercle Vaccine. The Serum was in each case heated to 60° C. for ten minutes.

Table supplementary to Table II.—*Showing that the Normal Serum, after it has been exposed to a Temperature of 60° C. for ten minutes, no longer incites Phagocytosis.*

		Unheated Serum.		Heated Serum.	
rial nber of ser- ion.	Nature of Infection.	Phagocytic Count. (Number of Bacteria ingested divided by Number of Leucocytes examined.)	Opsonic Index. (Determined by Comparison of Phagocytic Count with that obtained with Pooled Blood of Healthy Men.)	Phagocytic Count. (Number of Bacteria ingested divided by Number of Leucocytes examined.)	Opsonic Index. (Determined by Comparison of Phagocytic Count with that obtained with Pooled Unheated Blood of Normal Men).
1	Fibroid phthisis, tubercle bacilli in sputum	(100/30) = 3·3	1·0	(142/37) = 4·0	1·2
2	Early phthisis, tubercle bacilli in sputum	(132/30) = 4·4	1·3	(122/47) = 2·6	0·77
3	Acute phthisis, tubercle bacilli in sputum	(130/30) = 4·3	1·3	(96/40) = 2·4	0·74
4	Acute phthisis .	(127/40) = 3·2	1·0	(45/34) = 1·3	0·4
5	Fibroid phthisis (?)	(182/30) = 6·0	1·8	(51/43) = 1·2	0·3
6	Phthisis, tubercle bacilli in sputum	(117/30) = 3·9	1·1	(65/30) = 2·2	0·62
7	Mitral stenosis .	(106/30) = 3·5	1·0	(19/31) = 0·6	0·17
8	Early phthisis .	(161/30) = 5·4	1·6	(54/27) = 2·0	0·6
9	Phthisis . . .	(257/40) = 6·4	1·3	(51/40) = 1·3	0·27
10	Lupus under treatment by inoculation of tubercle vaccine	(131/36) = 3·3	1·6	(74/40) = 1·8	0·8
11	Lupus under treatment by inoculation of tubercle vaccine	(73/30) = 2·4	1·2	(31/30) = 1·0	0·5
12	Tubercular ulcer of leg under treatment by inoculation of tubercle vaccine	(63/30) = 2·1	1·2	(60/30) = 2·0	1·1

The first eight of the observations here in question were made upon bloods lected for us in the Victoria Park Hospital by our fellow-worker, Dr. G. W. Ross. e clinical diagnosis which had been arrived at was not made known to us till erwards, when the particulars set forth in Column 2 were filled in by Dr. Ross.

In contrast with the observations incorporated in Table II in t
body of the paper, these observations were conducted in phagocytic mixtu
containing 0·85 per cent. instead of 1·1 per cent. of NaCl. It is shov
in the next following communication that the spontaneous phagocyto
is absolutely abolished only in the case when the salt content of t
phagocytic mixture exceeds 1 per cent.

The source of fallacy to which attention is here called falls, no dou
or all practical purposes, entirely out of account.

On Spontaneous Phagocytosis, and on the Phagocytosis which is obtained with the Heated Serum of Patients who have responded to Tubercular Infection, or, as the case may be, to the Inoculation of a Tubercle Vaccine.[1]

By A. E. Wright and Staff-Surgeon S. T. Reid, R.N.

From the Laboratory of the Department for Therapeutic Inoculation, St. Mary's Hospital, London, W.

Recital of Previous Observations on the same Subject—Views of the Observers above-mentioned on the Nature of the Incitor Element contained in the Heated Serum—Sources of Fallacy which must be eliminated before the Question as to the Nature of the Incitor Element in the Heated Serum can be properly investigated—Fallacy of Spontaneous Phagocytosis—Investigation of the Influence of the Salt Content of the Fluid Environment of the Leucocyte upon Spontaneous Phagocytosis—Fallacy which may be introduced by the Exposure of the Serum for a Different Period to Different Degrees of Temperature—Investigation of the Question as to whether the Incitor Substance which is found in the Heated Serum of Persons who have responded to Tubercular Infection, or as the Case may be to an Inoculation of Tubercle Vaccine, is a Leucocytotropic Element—to which the Appellation "Stimulin" would apply—or Bacteriotropic Element, to which the term "Opsonin" would apply—Question as to whether the Opsonin found in Heated Immune Serum is or is not Identical with that found in the Unheated Normal Serum—Conclusions with respect to the Nature of the Incitor Element which is found in Heated Immune Serum after it has been Exposed to Heat.

It has been indicated in the foregoing paper that an *incitor element*[2] is to be found in the blood of those who have made an immunizing response to tubercular infection, or, as the case may be, to an inoculation of a tubercle vaccine. This fact does not stand by itself.

[1] Reprinted from the *Proceedings of the Royal Society*, B vol. lxxvii, 1906.

[2] The term "incitor-element" (Latin—*incito*: I hasten, I urge forward, I bring into rapid movement) is here employed to denote the element in the heated serum which promotes phagocytosis. By employing this term, pending the elucidation of the nature and mode of action of the element in question, we secure the advantage of leaving these issues unprejudged.

Recital of Previous Observations on the same Subject.

The observations of *Metchnikoff*, following in sequence upon th classical researches of R. Pfeiffer on the intraperitoneal destruction o bacteria by the aid of immune sera, first drew attention to the fact tha very active phagocytosis comes under observation when bacterial cultures or, as the case may be, spermatozoa, are introduced into the peritonea cavity of normal animals in association with heated serum derived fron immunised animals.

Savtschenko[1] obtained in experiments conducted *in vitro* with th heated sera of animals which had been subjected to injections of red bloo corpuscles, phagocytosis of these formed elements.

Neufeld and *Rimpau*,[2] working with heated sera derived from animal which had been immunized against streptococcus and pneumococcus and conducting their experiments *in vitro*, have described these immun sera as possessing a power of inciting phagocytosis. This power was, b it remarked, not numerically measured.

Leishman,[3] employing the numerical method for the measurement o phagocytosis which was devised by him with the modifications introduce by one of us in conjunction with Douglas, ascertained that the sera derive from Malta fever convalescents, or, as the case may be, from men wh had undergone anti-typhoid inoculation, retain, after heating, element which promote phagocytosis.

Dean, working with the same methods, without, however, conformin to the easily realized conditions [4] which are essential to the accuracy o the enumeration, has described incitor elements in the heated serun derived from animals which had been immunised against staphylococcus

Lastly, *Douglas*, employing again the same methods, has obtaine evidence of the presence of an incitor element in the heated serum derive from himself and others after inoculation with a sterilized culture of th plague bacillus.

Views of the Observers above mentioned on the Nature of the Incito Element contained in the Heated Serum.

Influenced by the theoretical conception that the increased resistanc

[1] *Annales de l'Institut Pasteur*, 1902.

[2] Neufeld and Rimpau's paper was published in the *Deutsche Med. Wochenschrif* in September, 1904, twelve months after the first description of the opsonins i these *Proceedings*.

[3] *Path. Soc. Trans.*, 1905, vol. lvi.

[4] "I should not feel disposed," remarks this author (*Roy. Soc. Proc.*, Serie B, vol. lxxvi, p. 511), "to place quite the same reliance as Wright and Douglas o the numerical accuracy of the results which can be derived from their method Where the leucocytes are very full, i.e., where the counts are high—it is impossibl to differentiate results by the method of enumeration." In spite of the perfectl self-evident experimental limitation of our method, which Dean here recognizes this worker employs in practically all his published experiments bacterial suspension which give him an *average* phagocytic count often of 50 and more bacteria in th leucocyte. Such a count is altogether incompatible with accurate quantitativ work.

bacterial invasion which is obtained by bacterial inoculation is in every se referable to a modification of the phagocytes,[1] Metchnikoff originly spoke of the incitor element as a *stimulin*.

This appellation may, we think, be characterized as unfortunate, *first*, cause the mode of action of the incitor element was prejudged; *secondly*, cause the appellation suggests (in contravention to everything which s come to light with respect to immunisation) that there are elaborated the animal organism in response to inoculations, not *vaccinotropic* ements (elements which have a chemical affinity for the vaccine) but *ucocytotropic* elements (substances which have a chemical action on ucocytes).

At a later date the terms "sensitizer" and "fixing substance" (*la bstance sensibilitrice* and *le fixateur*) were applied by Metchnikoff to e incitor element. This nomenclature is, it seems to us, almost equally felicitous—infelicitous because it imposes upon the mind the following eas:—(*a*) that the phenomena of phagocytosis are analogous to those haemolysis; (*b*) that the incitor substance, like the "amboceptor" Ehrlich, exerts its specific effect only in the case where it is reinforced a complement; and (*c*) that the mechanical movements of the phagocyte the ingestion of particulate matter are analogous to the chemical tion of the complement in the case where red blood corpuscles are ssolved by a haemolytic serum.

With the exception of Leishman,[2] who, with a view to conforming to e original nomenclature of Metchnikoff, and also because his own experients incline him to adopt the same point of view, speaks of the incitor bstances as *stimulins*, all the other observers[3] take the view that the

[1] The correctness of the view that artificial immunity depends upon a modifition of the leucocytes was first inquired into by Denys and Leclef (*La Cellule*, 95, vol. xi), in connexion with their experiments conducted on rabbits with eptococcus. The doubt with regard to the correctness of Metchnikoff's view ich found expression in the paper of these authors was further justified by the periments of Mennes (*Zeitsch. f. Hygiene*, 1897, vol. xxv), conducted with the blood animals immunised against the pneumococcus. Finally, the incorrectness of e view that immunisation depends on a modification of the leucocytes was for the st time unambiguously established by one of us working in conjunction with uglas (*Roy. Soc. Proc.*, vol. lxxii, p. 369, and vol. lxxiii, p. 129). Our results re afterwards confirmed by Bulloch (*Roy. Soc. Proc.*, vol. lxxv.)

[2] Loc. cit. and *Journal of Hygiene*, 1895.

[3] It may be remarked in this connexion that Neufeld and Rimpau, while satisd that the incitor substances in the serum exert an opsonic action on the bacteria, ggest that the term *opsonins* should be here rejected and that the substances re in question should be called *bacteriotropins*. Pending the discussion of the estion of the mode of action of the incitor elements in the heated serum, and their identity or non-identity with the opsonins found in normal blood, it will ffice here to remark with respect to the proposed nomenclature of Neufeld the lowing:—

(*a*) The term *bacteriotropins* (since it connotes nothing more than the property entering into chemical combination with bacteria) is more appropriate as a generic m for the whole class of substances which combine chemically with bacteria an as a specific designation for the substances which prepare the bacteria for agocytosis.

(*b*) All considerations of the comparative merits of Neufeld's terminology and

incitor element in the immune serum exerts an opsonic action upon th
bacteria, preparing them for phagocytosis.

Sources of Fallacy which must be eliminated before the Question as t the Nature of the Incitor Element in the Heated Serum can be properl investigated.

Before an inquiry into the nature of the incitor constituent of heate "immune serum" can be properly taken in hand, the sources of fallac which are incident to such an inquiry must be realized. A *first* source c fallacy is associated with the occurrence of *spontaneous phagocytosi* A *second* source of fallacy arises, as we shall see in a subsequent section, i connexion with the fact that the incitor power of the heated immur serum is influenced in a remarkable and, for the present, quite inexplica ble manner by the duration of the exposure to heat, and by the tempera ture employed.

Fallacy of Spontaneous Phagocytosis.

It will enforce itself upon the mind on considering the protocols c the original experiments published by one of us in conjunction wi Douglas[1] that the phagocytosis is not completely abolished by tl heating of even a normal serum. The residual phagocytosis registere in the protocols must, as reflection will show, be either *spontaneo phagocytosis*, meaning by this phagocytosis occurring apart from ar

my terminology apart—there must, I apprehend, remain to me as the author the term *bacteriotropic substances* (*Lancet*, December 23, 1899, p. 36 *supra*) as again Neufeld the right of assigning to this term its technical signification.

Dean likewise, while championing the view that the incitor element is an opsoni and while dissatisfied with the ambiguity of the terms "fixateur" and "substan sensibilitrice," and while conceding that "it may be convenient to adopt the ter *opsonin*," employs instead the periphrasis "*the substance which prepares the micr organisms for phagocytosis*," denying himself the convenience of the term opsoni "in order to mark the danger that one might be led to regard the opsonin as actual a different substance, and not merely a property of immune serum." My fello worker, Douglas, and I have not claimed for ourselves anything more than thi that we have, by the aid of an accurate quantitative method, adapted from Leishma placed in a clear light the rôle of the blood fluids in relation to phagocytosis, a r which was practically everywhere ignored or misconceived, and which had at be been "glimpsed" by one or two observers whose work, undertaken with ve defective and fallacious technical methods, was, as Dean's own analysis shows, a very unconvincing character. We submit that the clarification of the rôle the blood fluids which was effected by us would have remained incomplete a ineffective if we had not alighted on the terms "opsonic power" and "opsonins or some other apposite and equally convenient nomenclature to denote, as t case may be, the power or "*the substance in the serum which prepares the mic organisms for phagocytosis.*"

We would also submit that the ultimate—and we hold for the present una proachable question—as to whether the opsonic effect we have described is or one of a series of diverse effects exerted by a single *antitropic* substance, or wheth it is the result of the specific activity of an independent chemical unit in the seru is not prejudged by the employment of the term *opsonin.*

[1] *Proceedings of the Royal Society*, vols. lxxii and lxxiii, (pp. 75–99 *supra*).

co-operation of the serum, or phagocytosis dependent upon the chemical activity of an element which has resisted the destructive action of heat.[1]

When face to face with the consideration that the elimination of all spontaneous phagocytosis must be a necessary preliminary to the proper investigation of every question which has reference to the presence of an incitor element in heated serum, a suggestion from our fellow-worker, Captain Stewart R. Douglas, I.M.S., led us to inquire whether the phagocytic activity of the leucocyte might not be affected in a conspicuous manner by the salt content of its fluid environment. Captain Douglas's suggestion was a happy one. For, as will appear in the next section, we found that in certain concentrations of salt the leucocytes display considerable spontaneous phagocytosis with respect to the tubercle bacillus, while again in other salt concentrations spontaneous phagocytosis with respect to the micro-organisms is entirely suppressed.

Investigation of the Influence of the Salt Content of the Fluid Environment of the Leucocyte upon Spontaneous Phagocytosis.

The general results of our experiments conducted with tubercle bacilli will be best submitted in the form of the subjoined graphic curves.

In *Chart* 1 we show the phagocytic counts obtained in an experiment conducted without any admixture of serum. In these experiments one volume of washed blood corpuscles, suspended in 0·85 per cent. NaCl solution, was mixed in each case with an equal volume of suspension of tubercle bacilli in distilled water, and with one volume of a graduated solution of salt. It will be observed that the spontaneous phagocytosis which is here in question is greatest where the phagocytic mixture contains 0·6 per cent. of NaCl, and that the count falls off in a gradual manner, and finally reaches a figure which does not differ sensibly from zero when a concentration of 1·2 per cent. NaCl is arrived at.

In *Chart* 2 we show the phagocytic counts obtained in films prepared from phagocytic mixtures containing a *double* volume of undiluted serum, a *double* volume of washed corpuscles suspended in 1 per cent. NaCl, and a *single* volume of a suspension of tubercle bacilli made in the same menstruum, supplemented in each case by a *single* volume of a solution of sodium chloride of progressively increasing strength.

Three different sera were here subjected to experiment—

a. The pooled unheated serum derived from eight normal men;

b. The same serum after it had been exposed to a temperature of 60° C. for ten minutes; and

[1] In favour of the former of these two alternative explanations of the residual phagocytosis is, *first*, the difficulty of conceiving in connexion with the experiments conducted with carmine and Indian ink particles, that these were chemically acted upon by the serum; *secondly*, the difficulty of explaining, otherwise than as a result of individual differences in phagocytic activity as between leucocyte and leucocyte, the fact that in preparations made with heated normal serum and tubercle bacilli suspended in physiological salt solution, the phagocytosis is generally restricted to a very small percentage of the leucocytes instead of coming into evidence, as in the case of experiments conducted with unheated and active serum, in association with practically all the mature leucocytes.

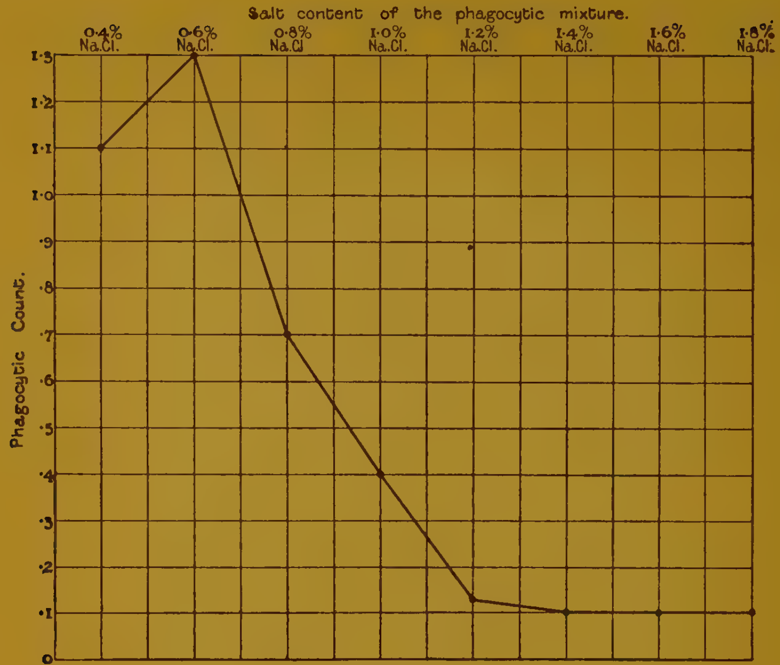


CHART 1.

c. Serum from a patient who had been subjected to therapeutic inoculations of tubercle vaccine. This serum, like the last, had been exposed to a temperature of 60° C. for ten minutes.

It will be seen that, as in *Chart* 1, where no serum was employed, the highest phagocytic counts were with each serum obtained where the concentration of the sodium chloride was least.

In the case of trace 3 (obtained with the heated normal serum) the

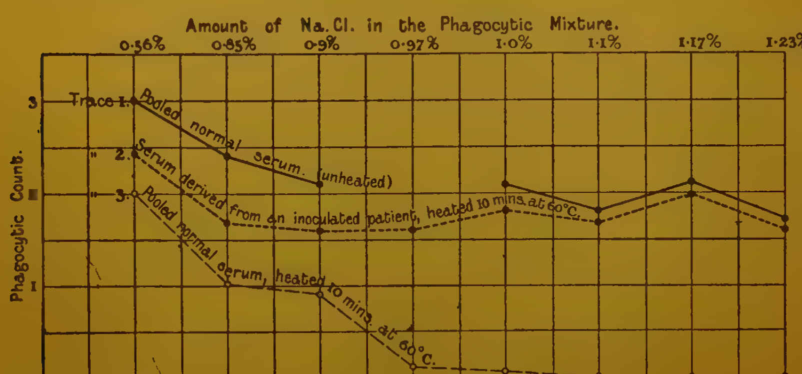


CHART 2.

hagocytosis must be interpreted throughout as purely spontaneous hagocytosis.

In trace 1 and trace 2 it must, in the case where low concentrations f NaCl are in question, be interpreted as spontaneous phagocytosis upplemented to an extent corresponding with the differences between the ounts in these traces and those in trace 3—by phagocytosis dependent pon the chemical action of the serum. Lastly, in these two first traces he phagocytosis registered where high concentrations of NaCl were em-loyed must be entirely dependent upon the chemical action of the serum.

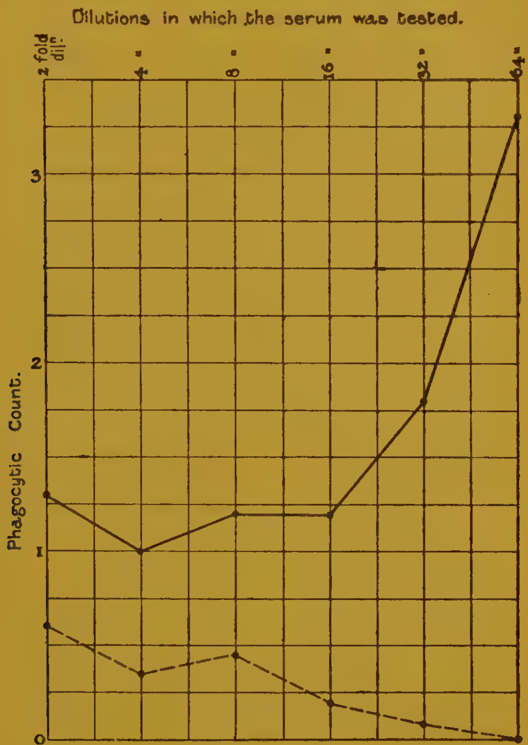


Chart 3.

In *Chart* 3 we show the effect of making progressive dilutions of one nd the same normal unheated serum with, in the one case, a 0·6 per cent. NaCl solution, and in the other case a 1·3 per cent. NaCl solution, using, s in the experiments above, in each case one and the same suspension of ubercle bacilli, and one and the same washed blood cream.[1]

It will be seen that while in the lower trace the phagocytic count ank away in an almost regular manner to zero as the opsonins of the

[1] By this procedure there were obtained in the first case phagocytic mixtures whose alt content diminished in the successive dilutions from 0·8 to 0·7 NaCl, and in the econd case phagocytic mixtures in which the salt content increased from 0·92 to 1 per cent.

serum were more and more diluted, in the upper trace the phagocytic count increased as the serum was diluted by a less concentrated salt solution.

We do not see room to doubt that in the case of the lower trace spontaneous phagocytosis was completely suppressed, and that such phagocytosis as was obtained was due exclusively to the action of the opsonins, and that in the case of the upper trace the phagocytosis obtained in the outset was due to spontaneous phagocytosis supplemented by the action of the opsonins, while the increased phagocytosis in the latter part of the trace was entirely due to spontaneous phagocytosis.

Fallacy which may be introduced by the Exposure of the Serum for a Different Period to Different Degrees of Temperature.

In view of the research of Dreyer,[1] which brought out the fact that the agglutinating power of a serum may, as progressively higher temperatures, or, as the case may be, progressively longer exposures are employed, be first lost and afterwards recovered, it suggested itself that an analogous effect might possibly be exerted upon the incitor element of an immune serum when exposed for different periods to different temperatures.

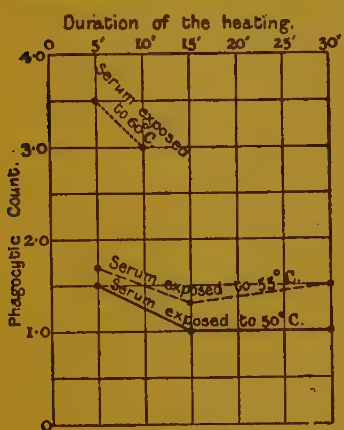


CHART 4.

The results of a typical experiment carried out with such a serum are embodied in the *Chart* 4.

It will be seen that, while the incitor power of the immune serum was in each case preserved, a very different phagocytic count was obtained according as the serum was exposed to temperatures of 50°, 55° and 60° for a shorter or longer time.

It will be manifest, in view of these results, that where experiments are conducted with heated sera care must be taken to see that in every case the same conditions are observed in the matter of the heating of the serum.

Investigation of the question as to whether the Incitor Substance which is found in the Heated Serum of Persons who have responded to Tubercular Infection, or, as the Case may be, to an Inoculation of Tubercle Vaccine, is a Leucocytotropic Element—to which the Appellation "Stimulin" would apply—or Bacteriotropic Element, to which the Term "Opsonin" would apply.

The absorption method of Ehrlich, which has already been employed by Neufeld and Rimpau in connexion with the investigation of the nature of the incitor element contained in the serum of animals immunised against streptococcus and pneumococcus, was obviously the method indicated for employment in connexion with the problem here before us. It was also

[1] *British Medical Journal*, September 10, 1904.

anifest, in view of the facts detailed in the previous section, that the mparative experiments instituted with heated immune serum before d after digestion with tubercle bacilli and subsequent centrifugalization ould yield unfallacious results only on adhering rigidly to the same nditions in the matter of the heating of the serum, and on arranging the perimental conditions in such a manner as to achieve in the phagocytic ixtures employed in each case a salt content of over 1 per cent. NaCl.

A series of experiments conducted with these precautions showed in uniform manner that the incitor element can be completely extracted om heated immune serum by digestion for half an hour at 37° C. with a spension of tubercle bacilli.[1]

It is thus clear that the incitor element which is found in heated serum persons who have responded to tubercular infection, or, as the case may , to the inoculation of a tubercle vaccine, is an opsonin. We may, nding the discussion in the next section of its identity and non-identity th the opsonin of the unheated normal serum, speak of this opsonin in a ovisional manner as the *opsonin found in the heated immune serum.*

estion as to whether the Opsonin found in Heated Immune Serum is or is not Identical with that found in the Unheated Normal Serum.

Leishman, who has spoken of the incitor element in the heated immune rum as a *stimulin*, in common with Neufeld, working in conjunction th Rimpau and Dean, who have shown that this incitor element nctions as an *opsonin*, have laid emphasis on the thermostability of the in- or element. Both Leishman and Neufeld urge that the character ther- ostability differentiates the incitor elements they have in view from the ermolabile opsonins described by one of us in conjunction with Douglas. eufeld goes further, and contends that the particular opsonins which ve been described by him as thermostable alone possess any significance connexion with the protection of the organism against bacterial disease. support of this contention Neufeld adverts to the fact that man, though he is, according to experiments recorded by one of us in conjunc- on with Douglas, the possessor of thermolabile opsonins against the ague bacillus, is none the less not protected against this micro-organism.

Before investigating the question of fact as to the identity or non- entity of the opsonins of the normal and immune organism, which are scriminated from each other by Neufeld, we may be allowed to comment the standpoint which he takes up. We submit that he proceeds upon entirely erroneous conception when he assumes that the non-immunised man organism does not offer a resistance to such bacterial infections plague. We submit, further, that it is erroneous to conceive of the rmal organism as differing from the immunised organism in a qualitative anner. Rather, does not the theory of Ehrlich brilliantly teach that in munisation we are never building upwards from a level of absolute

[1] This result is in conformity with the results obtained by Neufeld and Rimpau connexion with streptococcal and pneumococcal immune serum, and by Dean connexion with staphylococcal immune serum.

non-resistance, but always building upon a foundation which is alread laid—calling into existence in increased quantity and conveying into th blood only such chemical agents as exist already preformed in the body

Reverting from this digression, we may address ourselves to the inves tigation of the facts, and may inquire whether they plead against or favour of the identity of the opsonins which are found in the unheated no mal blood with the opsonins which are found in the heated immune blood

In the investigation of the facts we have built upon the followin postulates :—

(*a*) If the so-called thermostable opsonins are in reality thermostab it will make no difference to the result whether the serum heated in a diluted or in an undiluted condition. If, on the oth hand, the thermostable opsonins represent nothing other than residuum of thermolabile opsonins which has escaped destru tion by heat, it may quite well happen that the serum will completely inactivated if, before the heat is applied, the seru is adequately diluted.

(*b*) Again, if the serum as derived from an immunised organism co tains in its native condition a mixture of opsonins, which a respectively thermolabile and thermostable, we may, in confo mity with the all-round greater chemical stability of thermostab substances, expect that the thermolabile opsonins will be destroy when exposed to sunlight, and that the thermostable opsoni will remain unaffected.

(*c*) Lastly, if the reputedly thermostable opsonins constitute altogether new and distinct category of opsonins produced in t course of immunisation, we may expect, at any rate in cas where the immunisation has been carried very far, to find t thermostable opsonins greatly in excess of the thermolab opsonins. In such a case it would be reasonable to expect t heated serum to bear almost as much dilution as the unheat serum before the point is in each case reached where the opsor power is lost. On the contrary, if the so-called thermostab opsonins represent only an undestroyed residuum of the ordina thermolabile opsonins, we may expect the heated serum to forfe its opsonic power by dilution sooner than the unheated serum.

The graphic curves which are subjoined will serve to bring before t eye the results of, in each case, a typical experiment instituted with view to the resolution of the questions suggested above.

Chart 5 furnishes an answer to the questions suggested in (*a*) and (*c Chart* 6 an answer to the question suggested in (*b*).

Explanation of Chart 5.—The experiment, whose results are he graphically set forth, had a double object in view. Its first object w to determine whether the tuberculo-opsonic power of the serum deriv from an inoculated patient would be only partially abolished in the ca where heat is applied to the undiluted serum, and would be complete

plished when heat is applied to the diluted serum. Its secondary object s to determine how far one and the same serum could be diluted before d after heating before its tuberculo-opsonic power was extinguished.

The serum which was employed for the purposes of this experiment was tained from a patient whose opsonic index had been raised from 0·17 to by repeated inoculations of new tuberculin, and who had, under the luence of these inoculations, completely recovered from tuberculous

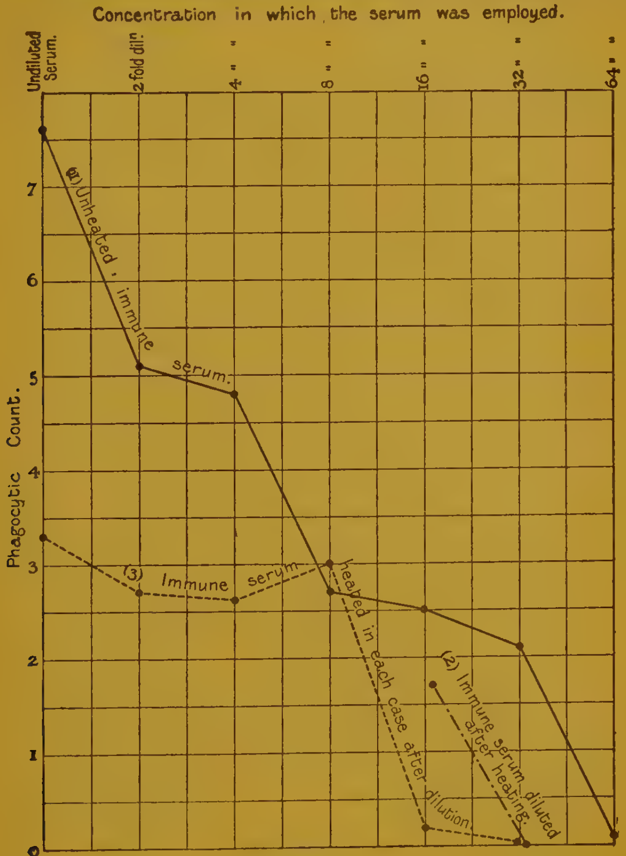


CHART 5.

ers of the leg, which had laid bare the tendons, and which had for a riod of thirteen years previously to the commencement of the inocu- ion treatment defied all treatment.

So far as the quantity of serum which was available allowed of this ng done, these questions were investigated, the precautions explained ove being in each case rigidly observed.

It will be seen on reference to the chart, and on comparison the phagocytic counts registered in the case of the 16-fold dilutions, at while the serum which had been first heated and then diluted (Trace

2) gave a phagocytic count of 1·7, the serum which had been first dilu and then heated (Trace 3) gave a phagocytic count of practically ze It will further appear on referring to the three traces that, while opsonic power of the unheated serum was maintained till a 64-f dilution was arrived at, the opsonic power of two samples of ser which were heated respectively before and after dilution extinguished when in the former case a 32-fold, and in the latter cas 16-fold dilution was arrived at.

Comment.—The experiment shows that the opsonin found in hea serum is destroyed by heat when the serum is sufficiently diluted.[1]

Explanation of Chart No. 6.—In the experiment here in question employed a serum derived from a patient with tubercular peritoni who had responded to infection in a characteristic manner.

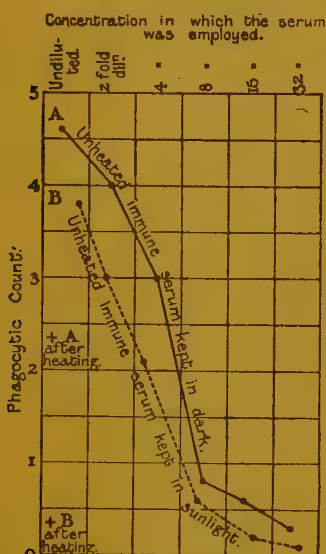


Chart 6.

Dividing it into two portions, we posed one portion to direct sunlight fo period of six to eight hours, keeping other portion in the dark in an incuba at 22° C. for the same time.

We now measured the opsonic pov of each portion of serum both in the heated condition and after exposure 60° C. for ten minutes. In the case the unheated samples we tested in ea case not only the undiluted serum but a in each case a series of progressive di tions. In the case of the samples which w heated we tested only the undiluted se

It will be seen on comparing the p gocytic counts obtained with the insola and non-insolated sera respectively, t while in the case of the unheated samp the serum which had been exposed sunlight gave throughout almost as hig phagocytic count as the serum which h been kept in the dark, in the case of the heated samples the serum wh had been exposed to sunlight gave a zero result, while the specimen wh had been kept in the dark gave a count of 2·3 bacilli to each leucocyte.

Comment.—The experiment shows that the reputedly thermosta opsonin is—in contradiction with what is known to hold of other th mostable elements—eminently heliolabile.

Conclusions with respect to the Nature of the Incitor element which found in Heated Immune Serum after it has been Exposed to Heat.

Manifestly the plain teaching of our experiments is, that the opso which is found in the heated immune serum of a patient who has respond

[1] Further experiments bearing on this question will be found in the Appen to the paper.

tubercular infection, or, as the case may be, to the inoculation of a tubercle ccine, does not differ with respect to its resistance to heat and sunlight om the opsonin which is found in the unheated normal serum.

A precisely similar conclusion with respect to the identity of the opson- s found respectively in unheated normal and heated immune sera was, may note, arrived at by Dean in connexion with his experiments on e sera of animals which had been immunised against staphylococcus.

We have only to remark in conclusion that if we prefer to speak of the sonin as a thermolabile element, and Dean prefers to speak of it as a ermostable element, there is nothing at issue between us except the estion as to whether it is in harmony with usage, and with the genius the English language as employed in scientific discourse, to charac- ise as "thermostabile" an element of which at best residual traces nain in the case of the normal serum where this has been heated to ° C., and in the case of the immune serum where this has, after equate dilution, been heated to the same temperature.

Appendix.

It may be convenient to subjoin here, in tabular form, the results of ee experiments, similar to that set forth in Curve 5 in which the opsonic wer of a tuberculo-immune serum was measured in a series of dilutions de in the one case after the serum had been heated to 60° C. for ten nutes, and in the other case before the serum was so heated.

Serial umber of he Experiment.	Source from which the Serum was derived.	Phagocytic Count obtained in the Case of the Heated Undiluted Serum.	Dilution in which the Opsonic Power was measured.	Phagocytic Count in the case where the Serum was heated before it was diluted.	Phagocytic Count in the case where the Serum was diluted before it was heated.
pt. 1 .	Pooled serum of six patients who had been inoculated with tubercle vaccine	2·4	2-fold dilution	1·9	3·3
			4-fold	2·7	1·7
			8- "	1·1	0·6
			16- "	1·0	0·45
			32- "	0·97	0·2
			64- "	0·75	0·08
pt. 2 .	Serum of a patient (E. M.) who had been inoculated with tubercle vaccine	—	4-fold	1·5	2·7
			8- "	1·4	1·9
			16- "	1·6	1·2
			32- "	1·5	0·3
				0·9	0·05
pt. 3 .	Serum of a patient (J. B.) who had been inoculated with tubercle vaccine	1·4	2-fold	—	1·5
			4- "	0·85	1·5
			8- "	0·7	1·6
			16- "	0·7	0·2
			32- "	0·25	0·0
			64- "	0·0	—

The Specificity of the Opsonic Substances in th
Blood Serum.[1]

By William Bulloch, M.D., and G. T. Western, M.A., M.B.

From the Bacteriological Laboratory of the London Hospital, London, E

A relatively high degree of specificity has been demonstrated for mo of the anti-bodies which exist in immune sera, e.g., in the case of agglutinin lysins, praecipitins, antitoxins. With normal sera the proof of specifici is often difficult on account of the fact that the antibodies are prese in the majority of cases only in small quantities.

The following experiments are concerned with the specificity of t opsonic substances of normal and immune sera. As is well known, the opsonic substances, discovered by Wright and Douglas, act on bacter in such a way that the latter become an easy prey to the phagocyt leucocytes.

If a given serum be tested it will be found to exert an opsonic acti on more than one kind of bacterium, and the question we have soug to answer is whether there is one or more than one opsonic substanc in other words, whether the opsonins are specific for the different bacte on which they exert their opsonic action.

In a previous communication [2] one of us (B) has shown that when microbe, e.g., staphylococcus, is digested with normal serum at 37° for fifteen minutes, and the cocci are then brought down by the aid of centrifuge, the supernatant liquid is found to be devoid of opsonic acti for staphylococci. Where the contact of the microbe with serum h been sufficiently long, and the centrifugalization has been complete, t opsonin for the particular microbe is totally removed.

We have attempted to determine whether the opsonins are speci by experiments of two kinds :—

1. The first method consisted in estimating the opsonic content of given serum towards two different bacteria. A suspension of one these bacteria was digested with the serum, and the mixture was thereaft centrifugalized, the resulting supernatant liquid being tested on bo kinds of bacteria. To a quantity of the supernatant liquid the secon

[1] Reprinted from the *Proceedings of the Royal Society*, series B, vol. lxxvii, 1906
[2] *Roy. Soc. Proc.*, vol. lxxiv (p. 138 *supra*).

acterial suspension was added, and after the lapse of a certain time the entrifuge was again applied, and the resulting liquid was again tested.

2. The second method consisted in estimating from day to day the psonic content of the serum of human beings suffering from lupus. At ertain periods tubercle or staphylococcus vaccines were inoculated, and e effect on the two opsonic curves was determined.

1. *Experiment on the opsonic action of normal human serum towards taphylococcus aureus and Bacterium pyocyaneum respectively.*

Normal human serum was mixed with an equal volume of a suspenon of *Staphylococcus aureus*, and the mixture was placed in the incubator r one hour at 37° C. At the end of this time the mixture was centrigalized, the supernatant liquid "A" being removed from the deposit cocci by means of a pipette. The supernatant liquid was in part tained, the remainder being digested for one hour at 37° C. with a ispension of *Bacterium pyocyaneum*, the latter being finally brought own as a deposit in the centrifuge, leaving a supernatant liquid "B," hich was pipetted off.

Result.

						Bacteria per Leucocyte.
1.	Normal serum	(1 in 2 dilution)	+ staphylococci	+ leucocytes	=	22·9
2.	,, ,,	(1 in 2 ,,)	+ *B. pyocyaneum*	+ ,,	=	4·7
3.	,, ,,	(1 in 4 ,,)	+ ,,	+ ,,	=	3·0
4.	Fluid "A"		+ staphylococcus	+ ,,	=	0·5
5.	,, "A"		+ *B. pyocyaneum*	+ ,,	=	4·0
6.	,, "B"		+ ,,	+ ,,	=	0·4

The contact of the serum with staphylococcus leaves the opsonic tion of the serum for *Bacterium pyocyaneum* practically unchanged, the yocyanic opsonin being finally removed by contact of the serum with is microbe.

A similar result was obtained when the serum was brought to act on aphylococcus and tubercle bacillus, as can be seen in the following periment.

(1) Normal human serum was mixed with an equal quantity of an nulsion of tubercle bacilli in 0·85 per cent. NaCl solution. The mixture as digested for thirty minutes at 37° C. and then centrifuged. In this ay a deposit and a supernatant liquid "A" was obtained.

(2) Normal human serum was mixed with an equal quantity of an nulsion of *Staphylococcus aureus* in 0·85 per cent. NaCl solution. The ixture was digested for thirty minutes at 37° C. and then centrifuged, supernatant liquid "B" being obtained.

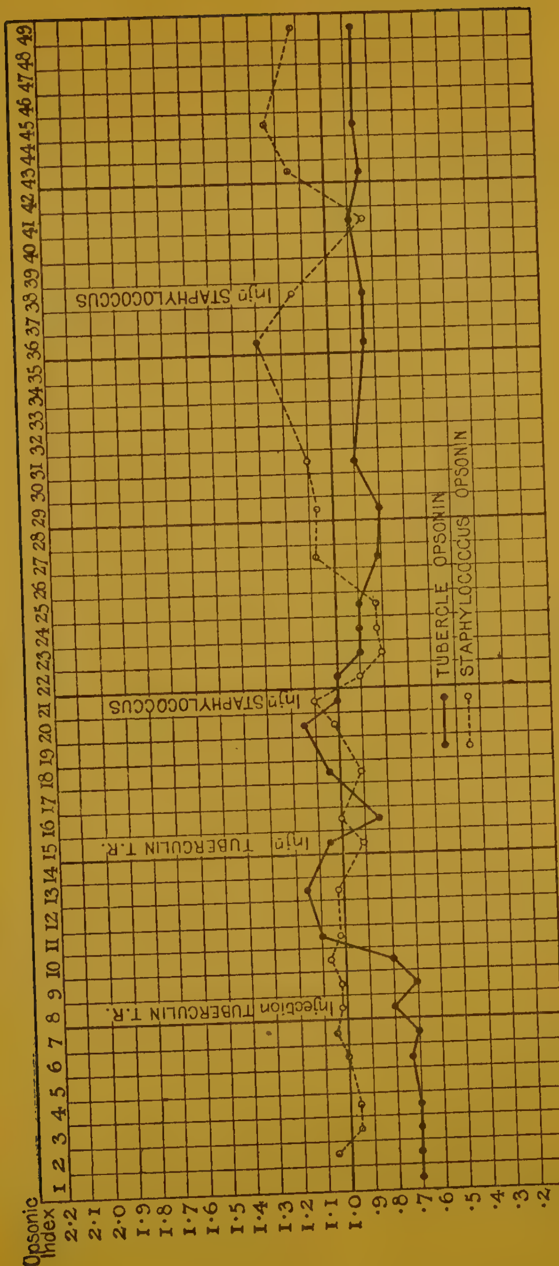
(3) The fluid "A" was mixed with an equal quantity of an emulsion *Staphylococcus aureus*. The mixture was digested for thirty minutes 37° C., and a deposit separated from a fluid "C" by the centrifuge.

4. The fluid "B" was mixed with an equal quantity of an emulsion of bercle bacilli. The mixture was digested for thirty minutes at 37° C., d a deposit separated from a fluid "D" by the centrifuge.

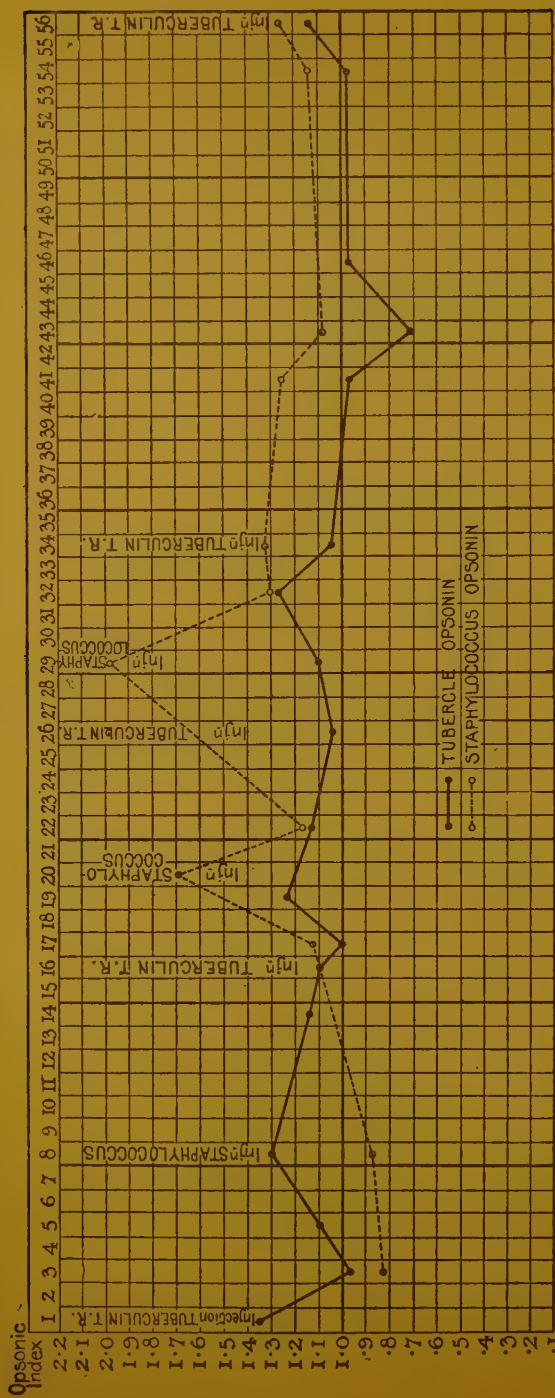
The opsonic content of the serum and of the fluids "A," "B," "C,"

Number of Microbes per Leucocyte.

	Expt. 1. Observer B.	Expt. 1. Observer W.	Expt. 1. Mean.	Expt. 2. Observer W.
1. Normal serum + saline *a.a.* (3 parts) + T.B. (1 part) + leucocytes (3 parts)	3·03	3·0	3·015	1·61
2. Normal serum + saline *a.a.* („) + staphylococcus („) + „ („)	12·6	12·3	12·45	7·00
3. Normal serum 1 + saline 3 („) + T.B. („) + „ („)	1·4	1·4	1·4	1·40
4. Normal serum 1 + saline 3 („) + staphylococcus („) + „ („)	11·0	11·4	11·2	5·20
5. Fluid "A." („) + T.B. („) + „ („)	0·4	0·5	0·45	0·13
6. „ "A" („) + staphylococcus („) + „ („)	9·0	10·23	9·96	5·00
7. „ "B" („) + T.B. („) + „ („)	2·7	—	2·7	1·20
8. „ "B" („) + staphylococcus („) + „ („)	0·43	0·26	0·34	0·80
9. „ "C" („) + „ („) + „ („)	0·13	0·26	0·19	0·40
10. „ "D" („) + T.B. („) + „ („)	0·16	0·76	0·51	0·32
11. „ "A" (3 parts) + saline (1 part) + leucocytes (3 parts) (stained for T.B.) . .	0·09	0·0	0·09	0·10
12. „ "B" („) + „ („) + „ („) („ for staphylococcus)	0·0	0·0	0·0	0·00
13. „ "C" („) + „ („) + „ („) („ „)	0·0	0·0	0·0	0·00
14. „ "D" („) + „ („) + „ („) („ for T.B.) . .	0·05	0·0	0·05	0·00
15. Saline, 0·85 per cent. (3 parts) + T.B. (1 part) + leucocytes (3 parts) .	0·0	0·08	0·08	0·06
16. „ 0·85 „ („) + staphylococcus („) + „ („) .	0·13	—	0·13	0·68


Opsonic Index
Injection TUBERCULIN T.R.
Injn TUBERCULIN T.R.
Injn STAPHYLOCOCCUS
Injn STAPHYLOCOCCUS
TUBERCLE OPSONIN
STAPHYLOCOCCUS OPSONIN

CURVE 1.


Opsonic Index
Injection TUBERCULIN T.R.
Injn STAPHYLOCOCCUS
Injn TUBERCULIN T.R.
Injn STAPHYLO-COCCUS
Injn TUBERCULIN T.R.
Injn STAPHYLOCOCCUS
Injn TUBERCULIN T.R.
Injn TUBERCULIN T.R.
TUBERCLE OPSONIN
STAPHYLOCOCCUS OPSONIN

CURVE 2.

d " D," was then determined both for *Staphylococcus aureus* and tubercle cillus in the usual way, the necessary controls being added. In the st experiments the determinations made by each of us separately in a ries of different films are given under the designation B. and W., and the ean of these determinations. In the second experiment the result was tained by one of us (W.) alone.

It will be seen that a considerable degree of specificity exists in so far at staphylococci remove almost the whole of the opsonin for this microbe, ile the opsonic substance for tubercle bacilli is in large part left unered. In almost all cases we have observed a slight diminution in the antity of the opsonin left behind. Thus, while the contact of a serum th tubercle bacilli lowered the opsonic content for this bacillus from)3 to 0·4, it also produced a slight lowering of the staphylococcus sonin from 11·2 to 9·96. Similarly contact of a staphylococcus with rum reduced the staphylococcus opsonin from 12·45 to 0·34, and at the ne time it lowered the tubercular opsonin from 3·0 to 2·7.

2. *Experiment.*

The opsonic content of the serum of a patient suffering from lupus was peatedly determined on tubercle and staphylococcus suspensions. Two oculations of tuberculin and two of staphylococcic vaccine were inted, and the influence of the inoculations is set forth in the following sonic curve, which shows that there is no correspondence in the quanies of tuberculo-opsonins and staphylococcus opsonins when one or ner of the corresponding vaccines is inoculated (Case I).

In a second experiment (Case II) opsonic determinations were made in imilar case, with the exception that the patient was not only suffering m lupus, but septic infection of the tuberculous lesions at the same ne (Curve 2).

Conclusions.

1. When staphylococci are brought into contact with normal human um and are subsequently removed by centrifugalization, the serum es its opsonic power for staphylococcus, although the opsonic power of *cterium pyocyaneum* is preserved.

2. Contact of normal human serum with tubercle bacilli leaves the sonic power of that serum for staphylococcus almost intact, while the sonic power for tubercle bacillus is completely removed.

3. Contact of normal human serum with staphylococcus leaves the sonic power of that serum for tubercle bacillus almost intact, while the sonic power for staphylococcus is completely removed.

4. Inoculation of a human being with tuberculin causes quantitative rease in the tuberculo-opsonin, whereas the quantity of staphylococcus sonin is unaltered.

5. Inoculation of a human being with staphylococcus vaccine causes quantitative increase in the staphylococcus opsonin, whereas the antity of tuberculo-opsonin is unaltered.

On the Relationship between Haemolysis and th Phagocytosis of Red Blood Cells.[1]

By R. D. Keith, M.A., M.D.

From the Bacteriological Laboratory of the London Hospital, London, E

Production of the Immune Serum which induces Haemolysis and Phagocytosis— Experiment to show that heating the Serum to 55° C. to 60° C. causes a Diminu tion of Phagocytosis—Behaviour of the Haemolytic Amboceptor towards He —Conclusions.

The nature of the substance or property in normal as well as in immun serum which induces phagocytosis has been of late a matter of considerab discussion, and the chief point of controversy has been whether phagoc tosis is caused by some well-known immune substance, or whether it brought about by something which until recently had not been complete recognized as a product of immunisation processes, e.g., the "opsonin" Wright and Douglas.

Whatever the nature of this subtance may be, it seems establishe beyond doubt that it acts on the bodies phagocytosed, the stimulin theo of Metchnikoff and his school having given way to the theory support especially by Wright and other observers in this country, that the actio is on the bodies phagocytosed and not on the phagocytes, notwithstandi the work of Löhlein (1), Leishman (2) and Besredka (3).

Wright and Douglas (4 and 5) in their well-known work on this subje described this property of the serum as being due to a body which up that time had not been properly recognized. To this they gave t name "opsonin," and by their ingenious experiments they rendered cle and concrete what had been before but nebulous and ill-defined.

They, as well as Bulloch and Atkin (6), and Hektoen and Ruediger (' described this body as being thermolabile from the fact that it was to large extent destroyed by heating the serum to 55° C. to 65° C. Dea (8) repeated this work, using a somewhat different technique, and havi found that in normal, but especially in immune sera, a certain amou was not destroyed, decided to call it thermostable. As Wright (9) h since pointed out, this is merely a matter of terms; but from his as w as from Dean's experiments it is clear that a very large amount of d struction takes place at these temperatures.

[1] Reprinted from the *Proceedings of the Royal Society*, series B, vol. lxxvii, 19

Dean at the same time put forward the view, shared chiefly by workers he Pasteur Institute in Paris, that the substance or property in the serum cribed by Wright and Douglas was not new, but had been well known ore, and Dean laid stress on the work of Savtchenko (10) on the phagocy-s of red blood cells, pointing out that this property had been attributed Savtchenko to the "fixateur."

As there seems to have crept into this question some doubt as to the ct interpretation to be put on Savtchenko's work, and particularly as he exact significance of the term "fixateur" as used by him, it is essary to briefly consider his position, especially as Barratt (11) has a different interpretation on it from Dean.

Savtchenko assumed that the laws regulating the action of cytotoxins e entirely analogous to those regulating the action of immunising stances on microbes, and considered that experiments on phagocytosis ht be permissibly conducted with animal cells and adopted red blood s, as being easy to work with.

This opinion would indicate that Savtchenko considered that the on of haemolysis was the analogue of that of immunising substances microbes, since a cytotoxic action with reference to red blood cells ld mean haemolysis. This is also indicated further on in his work n he says that, as has been pointed out by Bordet, when the red blood s of an animal A are injected into an animal B, the serum of the latter omes toxic for the red blood cells of the former, and that he himself established a complete analogy between the action of the serum on red blood cells and that of the immune specific serum on the microbe vell in the animal body as *in vitro*.

Further he says,[1] "Dans le sérum spécifique se trouve une substance fixateur (d'après la terminologie de Metchnikoff) qui se fixe sur les oules rouges correspondants—ou bien sur les microbes—et par son on prépare ces derniers à leur dissolution par les alexines (cytases) on trouve dans chaque sérum. Le fixateur ne se détruit pas à 55° C.)° C. Ehrlich et Morgenroth ont montré que le fixateur a une affinité cifique pour les globules rouges correspondants, et qu'une fois fixé sur , il ne s'en détache pas dans les lavages ultérieurs, ainsi que dans la trifugation dans l'eau physiolgique. Si l'on soumet les globules ges ainsi traités à l'action du sérum normal contenant des alexines, ils dissolvent."

With regard to Metchnikoff's definition of the fixateur which Savtchenko epts, one may state what Metchnikoff (12) himself has given in his est work on the subject.

On p. 355 he says, "Um in diesen bedeutungsvollen Ergebnissen das ier Festgestellte und das Hypothetische von einander zu halten, haben vorgeschlagen das Alexin oder Komplement unter dem Namen Cytase h. zellenlösendes Enzym), die sensibilisierende Substanz dagegen unter n Namen Fixator zu bezeichnen." He also states (p. 357) that Sav-

[1] Loc. cit., p. 111 (*vide references at end of paper*).

tchenko was the first to show that red blood cells which are laden with *specific fixateur* are extraordinarily easily phagocytosed.

Savtchenko stated further that he took as the objects of experim the phagocytes of the guinea-pig and its red blood corpuscles, and serum of a rabbit immunised against the red blood cells of the guin pig, and heated the serum of the rabbit to 55° C. to destroy the alexin leaving *the specific fixateur* intact. He also stated that he took the wash red cells of a guinea-pig and diluted them with normal saline solution a added a quantity of heated haemolytic immune serum in a dilution 1 in 200. After this mixture had been six hours at 37° C. he centrifugali and washed the corpuscles thrice with normal saline. "The red blo cells," he adds, "had attached to themselves *the fixateur*; since the ad tion of normal serum was sufficient to bring about the solution of haemoglobin."

Again,[1] he states, "Il est possible qu'il existe dans le plasma minimum de fixateur insuffisant pour être decélé par la réaction de disso tion, mais tout à fait suffisant pour provoquer la phagocytose après s'ê fixé sur ces derniers."

Savtchenko's position is this: As the result of his experiments came to the conclusion that in the serum of a rabbit immunised w guinea-pig's blood, there exists a substance which causes phagocytosis the red blood cells of the guinea-pig, and this substance, which n act either on the phagocytes or on the bodies to be phagocytosed, is specific fixateur, and possibly, according to the amount present in serum, this substance causes haemolysis or phagocytosis.

From what has been given here of Savtchenko's work it appears to beyond doubt that he considered that the specific fixateur which indu the phagocytosis of red blood cells is the same as the haemolytic ambocep of Ehrlich and not a separate body inducing this action.

Barratt[2] has shown that even with *unheated* immune serum, phagocyt of red blood cells may occur without the serum possessing either haemoly or agglutinative properties, and concludes from this that the phagocyt is not induced by the fixateur in the sense of the term as used by Savtcher nor by the agglutinin, but by some other body acting on the red blo corpuscles and not on the leucocytes. This body he placed in the class "opsonins."

Besredka (13), in summing up Barratt's paper, says, "Il y a, en ef dans un sérum haemolytique plusieurs substances. Est-ce le fixat (amboceptor) qui détermine la phagocytose ? est-ce l'agglutinine ? est enfin une troisième substance qui aurait uniquement pour fonction présider à la phagocytose ?" Besredka, it is clear, also assumes that fixateur is identical with the amboceptor.

The main question at issue, then, is whether the amboceptor, a by this I mean that acting in haemolysis, is identical with the substa

[1] Loc. cit., p. 118.

[2] Loc. cit.

ucing the phagocytosis of erythrocytes—the opsonin of Wright and uglas.

As Savtchenko and Barratt did not use exact quantitative methods in ir experiments, and as such are desirable, it has been necessary to use omewhat different technical procedure from that employed by these earchers, but the type of experiment was essentially the same as theirs.

Production of the Immune Serum, etc.

The materials used were the red blood cells of the ox, the serum of a bit immunised against these, and normal human leucocytes as the gocytic agents.

The rabbit received intra-peritoneally doses of 10 c.c. of washed ox puscles at intervals of a week, 30 c.c. in all being administered before eriments were commenced.

The last injection was made on November 13, 1905. On the 27th of same month it was found, testing in the usual way, that 0·002 c.c of serum produced, when fully complemented, total haemolysis of 2 c.c. 5 per cent. suspension in normal saline of washed ox corpuscles, after hours at 37° C. and twelve hours at room temperature.

the Effects of Heat on the Substances in the Serum which induces Haemolysis and Phagocytosis.

The first point to be studied was the influence of heat on the phagocytic on of the serum. With the *undiluted unheated* serum it was found to matter of considerable difficulty to perform phagocytic tests owing to molysis somewhat obscuring phagocytosis. With the *undiluted un-ted serum* only blood shadows were to be seen in the phagocytes, but diluting the serum sufficiently to suppress the effects of the complement, molysis was abolished and the red cells could be observed to be phagocy-ed, apparently in their normal condition.

In order to find approximately at what degree of dilution haemolysis uld cease to come into play, a series of haemolytic tests were performed capillary pipettes. This method was employed in preference to that inarily adopted, because with Wright's method of performing phagocytic s, to deal with absolute quantities is a matter of considerable difficulty.

Experiment.

Various dilutions of the *unheated* immune serum were made, and al parts of these dilutions and of a 5 per cent. suspension of the hed red blood cells of the ox were mixed in a series of capillary ettes, so that the ultimate proportion of serum in the mixtures varied n 1 in 2 to 1 in 100. These mixtures were placed at 37° C. for two rs. A parallel series was made with serum which had been heated 55° C. for fifteen minutes. This was placed in the same conditions as former series.

Dilutions of Serum in Mixtures.	Result.
1— 2	Complete haemolysis.
1— 6	Complete haemolysis.
1— 10	Marked haemolysis.
1— 20	Definite haemolysis.
1— 30	Trace of haemolysis.
1— 50	Trace of haemolysis.
1— 60	Haemolysis absent.
1— 70	Haemolysis absent.
1—100	Haemolysis absent.

This experiment shows that in the case of the *unheated* serum haemolysis took place in dilutions above 1 in 50, owing to dilution of t native complement and to the fact that no fresh complement was add With the *heated* serum there was no haemolysis, even with equal parts serum and of the suspension of corpuscles, although in such a diluti the *unheated* serum produced complete haemolysis.

It was therefore decided to begin phagocytic tests with dilutions abo 1 in 50 in the case of the unheated serum.

Experiment to show that heating the Serum to 55° C. to 60° C. caus a Diminution of Phagocytosis.

Unheated immune serum was diluted with normal saline solution the proportions of 1 in 15, 1 in 20, 1 in 30. Of each of these dilutions o part was mixed in a capillary pipette with one part of a 5 per ce suspension of washed ox corpuscles and one part of washed hum leucocytes, the final dilution being approximately 1 in 45, 1 in 60, 1 in The tubes were then placed for fifteen minutes at 37° C., films bei then made and stained with Leishman's stain.

At the same time series were made with portions of the serum whi had been heated to 55° C. and 59° C. respectively. The final dilutio in these were 1 in 3, 1 in 6, 1 in 12, 1 in 24, 1 in 45, 1 in 60. A contr consisted of one part of 0·85 saline, one part of the suspension of washed corpuscles and one part of washed human leucocytes.

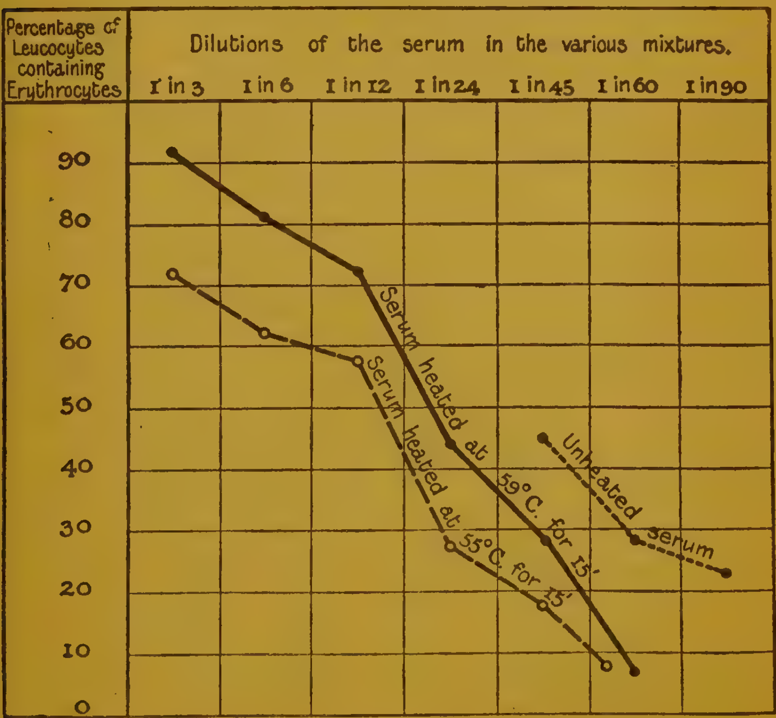
It was found in the first few dilutions that so many red blood ce were taken up by the polymorphonuclear leucocytes, that the individu erythrocytes could not be distinguished, and therefore the *percentage* polymorphonuclear leucocytes containing red blood cells was taken as criterion of the phagocytic action, 100 leucocytes being counted in ea case. Some of the large mononuclear leucocytes contained occasional one or two red cells, but these were so few as to be insignificant.

It was found that at corresponding dilutions the *unheated* seru produced a considerably greater amount of phagocytosis than did t *heated*, and further that it bore greater dilution before giving up th property.

Using the above-mentioned method of enumeration, the followi results were obtained in the experiment.

Dilution.	Unheated Serum.	Heated at 55° C.	Heated at 59° C.
	Per cent.	Per cent.	Per cent.
1 in 3 . .	—	71	91
1 in 6 . .	—	61	81
1 in 12 . .	—	57	72
1 in 24 . .	—	27	43
1 in 45 . .	45	18	28
1 in 60 . .	28	8	7
1 in 90 . .	23	—	—

Saline control=7 per cent. (The percentages refer to the number of ymorphonuclear leucocytes containing red blood cells.)



These results, which were confirmed by those of subsequent experi-nts, show that in an immune haemolytic serum the substance inducing agocytosis of the appropriate red blood cells is partially destroyed by ting the serum at 55° C. to 60° C. At the time of the experiment 02 c.c. of the serum when fully complemented produced complete emolysis of 2 c.c. of a 5 per cent. suspension of washed ox corpuscles er two hours at 37° C. and twelve hours at room temperature.

Behaviour of the Haemolytic Amboceptor towards Heat.

The next point for investigation was the influence of temperatur similar to those employed in the phagocytic tests, on the haemolyt amboceptor.

Experiment.

Of the immune rabbit's serum two portions were taken, one bei left unheated, the other being heated at 55° C. for fifteen minut Into two series of test tubes quantities of the serum ranging from 0· to 0·0001 c.c. were measured. One series then consisted of heate the other of unheated serum. All the tubes were equalized in bu by the addition of 0·85-per-cent. saline solution. To each tube 2 c of a 5 per cent. suspension of washed ox corpuscles were added, wi 0·2 c.c. of fresh normal guinea-pig's serum. One control consisted 2 c.c. of the suspension of red cells with 0·2 c.c. of guinea-pig serum, a another of 2 c.c. of the suspension alone. The tubes were placed 37° C. for two hours, and were subsequently allowed to remain twent four hours at 0° C. The corresponding dilutions in the heated a unheated series showed the same degree of haemolysis.

Result.

Unheated Serum.—Total haemolysis with all quantities down to 0·0 c.c. Partial haemolysis with all quantities down to 0·0001 c.c.

Heated Serum.—Total haemolysis with all quantities down to 0·005 c. Partial haemolysis with all quantities down to 0·0001 c.c.

In order to demonstrate conclusively whether there was any apprecia difference between the two series, von Fleischl's haemometer was employe the last three corresponding tubes in each series being compared with ea other and with the guinea-pig serum control. The tubes were thorough shaken up and centrifugalized. The supernatant fluid was then pipett off, and, if necessary, diluted sufficiently to give a reading between and 60 on the scale before being placed in the chamber of the instrume The reading found was then multiplied by the amount of the dilution.

The following are the results :—

	Colour Index.
Guinea-pig serum control . . .	64
Unheated serum—	
0·001 c.c.	440
0·0005 „	155
0·0001 „	120
Serum heated at 55° C. for 15 mins.—	
0·001 c.c	450
0·0005 „	220
0·0001 „	115

It is evident from these numbers that there is practically no differen between the colour indices of the two series; which permits the co clusion to be drawn that the haemolytic amboceptor is *not* quantitativ diminished when the serum is heated at 55° C. for fifteen minut

peated experiments gave exactly similar results, and it was found to a matter of indifference whether the serum was heated *en masse* or dilution, even in separate small quantities.

This fact is illustrated by the following experiment, which was per-med to illustrate at the same time another point, namely, that there y be a large amount of haemolytic amboceptor present in a *diluted* um without the co-existence of the body inducing phagocytosis.

Experiment.

Four series of tests, A, B, C, D, each consisting of four tubes, were rformed. Into successive tubes of each series 0·01, 0·005, 0·003, 0·002 . of the immune serum was placed. The amounts were equalized by 35 saline solution. Series A and C were unheated. Series B and D re heated at 55° C. for fifteen minutes. To each tube was then added c.c. of a 5 per cent. suspension of washed ox corpuscles, and to each be of series A and B 0·2 c.c. of fresh guinea-pig serum (i.e., one unheated, d one heated series was complemented). All the tubes were placed at ° C. for two hours. It was then found that series A and B showed actly corresponding degrees of haemolysis.

Cubic Centimetres.	A. Unheated and Complemented.	B. Heated and Complemented.
)·001	Almost complete	Almost complete
)·005	Marked	Marked
)·003	Slight	Slight
)·002	Slight	Slight

This first part of the experiment corroborates the result of the experi-nt mentioned immediately above.

In series C and D (the proportions of the serum in the mixtures rresponding to 1 in 220, 1 in 450, 1 in 730 and 1 in 1,100 approximately), e series being heated and the other unheated, and both being uncom-mented, it was found that there was no sign of haemolysis when these re compared with the controls, which were the same as in the previous periment. The tubes of these two series were thoroughly shaken d centrifugalized. The supernatant fluid was pipetted off and the posits washed thrice with 0·85 saline solution. To each deposit an ual quantity of normal saline was added. They were then well shaken d drawn up and down rapidly in capillary pipettes in order to produce uniform suspension. Equal parts of each deposit and washed human icocytes were mixed in capillary pipettes and placed fifteen minutes at ° C., films being then made and stained in the usual manner.

Result.—In no case was any phagocytosis observed, although in dilu-ons of 1 in 10 similarly treated, 93 per cent. of the polymorphonuclear icocytes contained erythrocytes, which shows that such deposits can phagocytosed, provided that the substance which induces phagocytosis present in sufficient amount. Although in series C and D no phagocytosis

occurred, yet in dilutions of 1 in 220 haemolysis was almost complete the complemented series, which shows that there must have been a la amount of haemolytic amboceptor present, and that notwithstanding t large amount of amboceptor and an exposure during two hours of red blood cells to it, no phagocytosis was observed.

This second part of the experiment then shows that in an immu *diluted* haemolytic serum a considerable amount of haemolytic aml ceptor may be present without rendering the red cells capable of bei phagocytosed.

This is supported by observations on non-immune haemolytic sera. the case of a guinea-pig's serum which was found in dilution of 1 in 6 produce slight haemolysis of 2 c.c. of a 5 per cent, suspension of the wasl blood corpuscles of a rabbit, it was observed that in phagocytic te performed with the unheated serum, the human leucocytes used as phagocytic agents contained in many cases blood shadows. These w found in 40 to 50 per cent. of the leucocytes in tests performed in manner described in the former part of this paper. When, howev heated serum is employed no blood shadows are to be seen in the leu cytes nor is there any sign of phagocytosis.

In this case of the serum of an eel it was found that 0·01 c.c. produ after two hours at 37° C. marked haemolysis of 2 c.c. of a 10 per ce suspension of washed guinea-pig red cells. When heated at 55° C., howev such a serum failed to induce phagocytosis of the red cells after 15 minu at 37° C., equal parts of the serum, of the suspension of red cells and washed human leucocytes being employed.

All these facts, then, tend to show that the haemolytic amboceptor m be present in a very considerable amount in a serum without giving to latter the power of inducing phagocytosis of the appropriate red blo cells.

Conclusions.

The conclusion naturally come to is that the phagocyto of red blood cells does not depend on the presence of the haemoly amboceptor, since :—

1. The substance which induces phagocytosis is partially destroy by heat, while the haemolytic amboceptor is entirely thermostable.

2. The haemolytic amboceptor may be present in considerable amou in a haemolytic serum without inducing phagocytosis, notwithstandi prolonged contact of the amboceptor with the red blood cells. This contrary to the opinion of Savtchenko.[1]

Dean [2] has suggested that phagocytosis may be caused by a comp ment acting through an amboceptor, and that the partial destruction the property in the serum inducing phagocytosis by heat may be due the destruction of the complement, while the amboceptor, even in absence of the complement, may still be capable of inducing phagocyto

[1] Loc. cit., p. 118. [2] Loc. cit.

his theory, while it is difficult to disprove directly owing to the comple-
ient being destroyed at the same temperature as the thermolabile part f the substance inducing phagocytosis, seems to be an improbable one, or the following reasons :—

(1) That it is not an action analogous to that of other amboceptors, g., that concerned in haemolysis. If one destroy the complement of a aemolytic serum by heat, no haemolysis takes place, notwithstanding the resence of the amboceptor in large amount.

(2) As has been shown above, the haemolytic amboceptor may be resent in large amount in a diluted serum, without that serum having ie power of inducing phagocytosis, even when Dean's method of testing employed.

(3) In the dilution experiments recorded above it was shown that one iay dilute the complement to such an extent as to abolish haemolysis, nd yet such a serum has a greater "opsonic" power in these dilutions ian has the same serum when heated and employed in corresponding ilutions.

If the amboceptor act in the way Dean suggests, it must be supposed possess, in addition to its complementophilic group, another group hich possesses the special function of inducing phagocytosis, i.e., the nboceptor would combine the functions of the second and third receptor ypes of Ehrlich.

The experiments given in this paper, along with those of Barratt,[1] nd to show that, contrary to the opinion of Dean, Savtchenko was ot correct in his conclusion that the specific fixateur, i.e., the haemolytic nboceptor, induced the phagocytosis of red blood cells, but that, on ie other hand, it is much more probable that this phenomenon is caused y some special body belonging to the class of opsonins.

I have to thank Dr. F. W. Twort for performing the experiments on iimals. I have also to express my thanks to Mr. J. A. Craw for sug-
estions, and to Dr. W. Bulloch, of the London Hospital, for his kind lvice and assistance during the course of my work.

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Part II

ON THERAPEUTIC IMMUNISATION

Notes on the Treatment of Furunculosis, Sycosis, and Acne by the Inoculation of a Staphylococcus Vaccine;

nd generally on the Treatment of Localized Bacterial vasions by Therapeutic Inoculation of the corresponding Bacterial Vaccines.[1]

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[1] Reprinted from the *Lancet*, March 29, 1902.

Introductory

The circumstance that inoculation against pyogenic bacteria has no up to the present, been resorted to would seem to be due to the fact tha our range of thought is in this, as in every other direction, limited by pr suppositions. The following are among the pre-suppositions which a current in connexion with inoculation.

(1) Since methods of inoculation find their special sphere of applica tion in connexion with septicaemic diseases there would be litt prospect of a useful application of such methods in connexion wit any local invasion of the tissues by pyogenic bacteria.

(2) Since pyogenic micro-organisms can always be held off fro operation wounds by a rigid observance of aseptic precautions, the would be no prospect of a useful application of inoculations such as w have here in view in connexion with operative procedures.

(3) Since bacterial invasions of the skin and mucous membran can be dealt with by the application of chemical antiseptics a resort inoculations directed against the invading bacteria would here also b out of place.

(4) Inoculations with vaccines can be of avail only when the forestall infection.

(5) Where a vaccine containing toxic bacterial products is introduce into the organism of a patient who is already infected this mu inevitably be followed by an aggravation of that patient's condition.

Let us take the above assumptions in order and see how far each them is justified.

The first assumption can be disposed of in a few words. is, in point of fact, merely the expression of the belief of a peri when inoculation—using the term in its widest sense—had as yet be applied only in connexion with the prophylaxis of small-pox. Sin the period in question methods of inoculation have, as reflection will sho been successfully applied in connexion with almost every form of bacteri invasion.

In connexion with the second assumption it will suffice to rema that, even assuming that inoculations against pyogenic bacteria wou fail to find any useful application in connexion with operative procedur such inoculations might conceivably render service in connexion with ba terial invasions of the skin and mucous membranes.

The next assumption—to wit the assumption that bacterial invasio of the skin and mucous membranes can be dealt with by the application chemical antiseptics—altogether ignores the defects and limitations of th method of treatment. These defects and limitations—already fully realiz in connexion with the treatment of operation wounds—are (*a*) the injury the tissues inflicted by the antiseptic; (*b*) the difficulty of destroyi bacteria which have penetrated below the surface; (*c*) the difficulty applying the antiseptic over the whole affected area in such a manner

make an end once for all of the invading bacteria ; and (*d*) the diffi-
lty of preventing the re-invasion of the disinfected from the neigh-
uring still infected areas.

Let us note that, assuming that it were found possible to inoculate
such a manner as to call into action the defensive powers of the
ganism, all these difficulties would have been effectually overcome.

We next pass on to consider how far we can uphold the assumption that
oculations with bacterial vaccines must be unavailing unless where they
restall infection, and the further assumption that inoculations of
erilized bacterial cultures—i.e., inoculations which involve the intro-
ıction of toxic bacterial substances into the organism—are necessarily
sociated with risk when undertaken upon patients who are already the
bjects of the corresponding bacterial infections.

Consideration shows that these assumptions express the results of ex-
erience derived from an observation of the effect of inoculations under-
ken in the case of animals and men when already affected with
pticaemic diseases. Confining ourselves to the case of man and to
e case of inoculations undertaken with sterilized bacterial cultures,
ere would, indeed, seem grounds for believing that an aggravation
the patient's condition has in some instances followed in the case
here the patient has been inoculated with anti-typhoid [1] and anti-
ague [2] vaccine respectively while in the incubation stage of these
seases.

But we cannot generalize from such instances.

For I have in a previous communication,[3] dealing with the changes
fected in the blood by anti-typhoid inoculation, pointed out that
ere is an all-important difference between the effects produced re-
ectively by large and small doses of vaccine. It was shown in the
per just referred to (*a*) that where the dose of bacterial vaccine employed
as sufficient to produce marked constitutional symptoms, inoculation
as followed, first, by a negative phase of diminished bactericidal power
-corresponding, it may be presumed, to a phase of diminished resistance
-and, then, after an interval of a few days, by a positive phase of increased
ıctericidal power—corresponding, it may be presumed, to a phase of
creased resistance ; (*b*) that where the dose of vaccine was such as to
oduce only very slight constitutional disturbance the negative phase
as suppressed, the positive phase being already distinctly marked
ithin twenty-four hours after inoculation ; and (*c*) that where the dose
vaccine was such as to produce very considerable constitutional dis-
ırbance the negative phase was exaggerated and prolonged, while the
ositive phase was considerably, possibly indefinitely, postponed.

Since an essentially similar succession of a negative and a positive

[1] Wright, *The Lancet*, September 14, 1901, p. 715, and *Brit. Med. Journal*, Oct.
, 1901.

[2] Report of the Indian Plague Commission, chapter iv, p. 195.

[3] *The Lancet*, September 14, 1901, p. 715.

phase manifests itself after the inoculation of the toxins of both tetanus and diphtheria, and, as we shall see below, also in connexion with the inoculation of staphylococcus vaccine, and since the extent and duration of the phase of diminished resistance are in each case a simple function of the dose administered, it would seem clear that the doctrine of the necessary inefficacy and invariable risk of inoculations undertaken upon patients already infected must be abandoned.

Manifestly, inoculation will be associated with risk only where the dose of vaccine employed is such as seriously to diminish the patient's power.

It may be noted here that Mr. Haffkine has from the outset ascribed to his anti-plague vaccine a power of aborting and diminishing the severity of an attack of plague in the case of any patient who may have been inoculated in the incubation stage of the disease. Mr. Haffkine is entitled to the fullest recognition of the fact that in certain of the cases recently recorded by Miss Corthorn,[1] and, it may be presumed, in other cases included in the statistical table published by Major W. B. Bannerman, I.M.S.,[2] the event would appear to have been quite in accordance with Mr. Haffkine's anticipations.

Leaving out of consideration all incidental issues, such as that of the expediency of inoculating in the incubation stage of any septicaemic disease unless with duly reduced doses of a standardized vaccine, let us here concentrate our attention upon the fact that it would seem possible —given the employment of the appropriate dose of a vaccine and given also certain other conditions presently to be considered—to obtain benefit from bacterial inoculations even in the case where the patient is already the subject of bacterial infection.

If it holds true that inoculations with bacterial vaccines may upon occasion have a therapeutic value in the incipient stages of bacterial invasions, which may afterwards assume a septicaemic form, it must *a fortiori* hold true that inoculations conducted with appropriate doses of bacterial vaccines may render useful services in the case of bacterial invasions which manifest themselves from first to last only in the form of localized inflammatory processes.

For—and this is clearly brought out by a comparative study of bacterial disease in animals, as well as by the study of any series of cases of one and the same bacterial infection in man—the localization and restriction of a bacterial invasion may always be taken as an indication of relatively high grade of resistance on the part of the infected organism.

If we consider the matter rightly, we shall see that the process of bacterial inoculation as applied to a patient who is the subject of a bacterial invasion is, in reality, a process of temporarily taking away from patient's power of resistance with a view to his receiving back that power with usury.

It is, in short, a process of trading upon the patient's balance o

[1] *British Medical Journal*, January 25, 1902. [2] Ibid., September, 14, 1901.

stance. When exploiting the method it will, then, be wise to inform selves beforehand, concerning the resisting power of the patient. It be wise, also, before adventuring, to learn what we can about the ands which will be made upon the patient's resisting power by the ulation of that definite quantum of the bacterial vaccine which we oose to employ.

Having now, so far as I can, made clear the situation, I pass on to set h the results of some preliminary work recently undertaken in con-ion with the therapeutic application of anti-staphylococcus inoculation he treatment of localized staphylococcus invasions. The vaccines loyed consisted in each case of staphylococcus cultures which had n sterilized by heating. The cases, six in number, which are reported w, include all that I have treated by this method.

CLINICAL AND BACTERIOLOGICAL DATA RELATING TO CASE I.

The patient, a man 40 years of age, consulted me in September, 1900, n a view to seeing whether anything could be done for him. He had ered from furunculosis, complicated by sycosis and eczema of the , since 1893. In that year, while engaged in clearing out a trache-my tube which had been removed from a patient who had been oper-l upon for acute laryngitis, he accidentally inoculated himself in the finger with some of the septic material. In spite of three deep sions made successively into the finger and the palm of the hand, ction spread upwards to the axilla (giving origin there to a bubo) thence onwards into the blood-stream, setting up high fever and septic tonitis. Since that time the patient, who had not previously suffered his way, had been afflicted with the disorders referred to above. ing the seven years which had elapsed since the onset the patient ared that he had never been free from boils for more than three months secutively. Change of climate, every form of medicinal treatment, local applications of the most varied description had all failed to rd any permanent relief. The patient was now suffering from an cerbation of his symptoms. Recently he had hardly ever been quite from styes on the eye, and boils. These last apparently occurred in varieties—small superficial boils taking their origin in the skin on the k and face, and larger deep-seated boils occurring in the subcutaneous ue in any and every region of the body. The condition of sycosis associated with weeping eczema, and was best marked on the region he chin. The infiltration of the deeper layers of the skin was even there tively inconsiderable. The hair follicles of the eyelashes and eyebrows e affected by an inflammatory process similar to that which was affect-the hair follicles of the beard.

Data of the bacteriological examination of the contents of the boils.—cultivation, made on September 16, 1900, from a small superficial on the face yielded a pure cultivation of the staphylococcus albus.

A cultivation made on September 19 from a larger and more deep-sea
boil on the forehead yielded a pure cultivation of the staphylococcus aure
A cultivation made on the 24th from a large boil on the forearm yielc
a pure cultivation of the staphylococcus aureus.

Data of the blood examinations instituted before inoculation.–
count of the different varieties of white corpuscles showed these to
present in the blood in normal proportions. A measurement[1] of
alkalinity of the serum showed a normal degree of alkalinity : $\frac{N}{35}$.

Estimation of the inhibitory power exerted by the serum on the grov of the staphylococcus.—Comparative counts[2] were made of the numbe staphylococcus colonies which developed in a series of measured volur of gelatine culture mixed (*a*) with sterile broth, (*b*) with serum deri from normal men, and (*c*) with serum derived from the patient. I somewhat extensive series of observations—carried out on three samp of the patient's blood and upon 13 control samples of normal blood dra off from six normal persons—only inconstant and very inconsidera differences were found between the number of colonies developing in tubes filled in with three different admixtures particularized above. are thus entitled to conclude that neither the serum of the patient any of the control sera exerted any inhibitory effect upon the grov of the staphylococcus.

Investigation of the agglutinating power exerted by the serum upon staphylococcus.—This was undertaken by mixing together in capill tubes equal volumes of a suspension of an agar culture of staphylococ and serum in various dilutions. The observations were underta on three different samples of the patient's blood. On one occasion o was a sedimentation obtained, and this was obtained only in the two-f dilution of the serum. The sera of four normal men were at the same ti examined by the same method. Two of these showed an agglutinat reaction in all dilutions up to a 16-fold dilution. The two others sho the reaction only in the two, four, and eight-fold dilutions.

Preparation of the staphylococcus vaccine which was employed in treatment of the patient.—Nutrient broth was inoculated with a staph coccus aureus which had been subcultured from one of the original cult obtained from the patient. After cultivation for three weeks at a temp ature of 37° C. the culture was sterilized by exposure for 20 minutes t temperature of 65° C. An addition of 0·5 per cent. lysol was made to sterilized culture. The toxicity of the vaccine was then tested on guin pigs. It was found that when the quantum of the vaccine inocula corresponded to 2 per cent, of their body-weight a moderate amount oedema and a certain amount of constitutional disturbance were produc

[1] The technique employed was that described by me in *The Lancet* of Septem 18, 1897, p. 719.

[2] The technique employed was that described by me in *The Lancet* of Decem 1, 1900, p. 1556.

Details of the inoculations carried out upon the patient, and resulting ...ical symptoms.—On October 25, 1900, the patient was inoculated ...cutaneously in the flank with one cubic centimetre of the above ...cine. Twelve hours afterwards there was considerable headache, the ...ient's temperature stood at 100·6° F., and considerable redness and ...derness had developed at the site of inoculation. The symptoms had ...irely passed off after the expiration of 36 hours. On November 1 1·5 ...ic centimetres of the same vaccine were inoculated. The inoculation ...s followed by similiar but somewhat less severe local and constitutional ...nptoms. On the 13th 1·5 cubic centimetres of the same vaccine were ...culated. Both the local reaction and the general reaction were very ...ht, the temperature not rising above 99·4° F.

Data of Blood Examinations instituted in the Period supervening upon ...**culation.**—*Investigation of the agglutinating power exerted by the serum* ...*on the staphylococcus.*—A determination of the agglutinating power in-...uted subsequently to the second inoculation showed that the serum now ...glutinated the staphylococcus in all dilutions up to a 16–fold dilution.

Investigation of the growth-inhibiting power exerted upon the staphy-...occus.—An estimation carried out in the interval between the first ...l second inoculations showed that the serum had not acquired any ...ver of inhibiting the growth of the staphylococcus.

The results of two successive estimations of the growth-inhibiting ...ver instituted soon after the dates of the third inoculation are subjoined ...tabular form (Table I).

...BLE I.—*Estimation of the Growth-inhibiting Power exerted upon the Staphylococcus Aureus by the Blood of Patient* 1 *soon after his Third Inoculation.*

...ate on which ...he Blood was examined.	Number of colonies of Staphylococcus which developed in the Capillary Tubes filled in each case with 5 cmm. of a Gelatin Culture of Staphylococcus (previously diluted 100,000-fold and 1,000,000-fold respectively with nutrient gelatin) and				Number of Colonies of Staphylococcus which developed in Control Tubes filled in each case with 5 cmm. of same Dilution of gelatin culture and 5 cmm. of sterile broth.		
	5 cmm. of Patient's Serum.	5 cmm. of a 2-fold Dilution of Patient's Serum.	5 cmm. of a 4-fold Dilution of Patient's Serum.	5 cmm. of an 8-fold dilution of patient's serum.	Tube 1.	Tube 2.	Tube 3.
...v. 14, 1900 .	0	2	5	10	57	22	50
19, „ .	8	7	7	10	29	36	23
...erage . .	4	4·5	6	10	36·1		

The results of the estimations set forth below would seem to indicate ...at the blood of a patient in the period immediately subsequent to inocu-...ion constituted a somewhat unfavourable medium for the development

of the staphylococcus. The question as to whether any actual bacte dal action was exerted was unfortunately not determined.

Clinical history of the patient subsequently to inoculation.—From date of the first inoculation in October, 1900, the patient's condition be to improve. With the exception of two small superficial boils which veloped between the dates of the second and third inoculations and further similar small boils which developed in the earlier part of 1 the patient has been absolutely free from furunculosis. The syc eczema, and the affection of the eyelids also began to mend from the c of the first inoculation ; the two former had practically disappeared wi a month from the beginning of the treatment. The ophthalmia t lasted a little longer. His face is now, and has for a period of ove months been, absolutely clean and free from eruption.

Data of blood examinations instituted more than a year after the dat the original inoculation.—Recently the patient's blood has been exami by the method devised by my colleague, Major W. B. Leishman,[1] measuring the phagocytic power of the blood *in vitro*. The result these estimations are subjoined in tabular form (Table II).

TABLE II.—*Results of the Estimations of the Phagocytic Power of Blood of Patient* 1 *instituted more than* 12 *months after the Dat Inoculation.*

Date on which the Blood was tested.	Source and Variety of Staphylococcus employed.	Average Number of Staphylococci ingested by each Polynuclear White Blood Corpuscle of the *Normal blood.*	Average number of staphylococci ingested by each polynuclear white blood corpuscle of the *Patient's* blood.	Phagocytic Inde i.e., the Proporti in which the Num of Staphylococ ingested by Pol nuclear White Bl Corpuscles of t *Control* blood st to the Numbe ingested by Polynuclear Wh Blood Corpuscle the *Patient's* blo
1901. December 2 .	Staphylococcus aureus subcultured from Patient 1	9·3	21·7	1 : 2·3
„ 15 .	Ditto	6·5	9·5	1 : 1·45
„ „ .	Staphylococcus albus subcultured from Patient 3	12·2	12·9	1 : 1·07
1902. January 23 .	Ditto	32·8	35·7	1 : 1·07

The control blood employed in the three first-recorded estimations was deri from W. B. L. : in the last estimation it was derived from A. E. W.

It will be manifest that the capacity of the patient's white bl corpuscles for ingesting staphylococci—and more particularly for ingest

[1] *British Medical Journal*, January 11, 1902.

e variety of staphylococcus with which he had been inoculated—was in ch case found to be greater than that of the white blood corpuscles of e normal blood used as a control. These observations—more especially en taken in association with the observations made in connexion with e cases recorded below—lend probability to the assumption that the tient's continued freedom from staphylococcus invasion is the result the inoculations undertaken.

CLINICAL AND BACTERIOLOGICAL DATA RELATING TO CASE 2.

The patient, an elderly maiden lady, presented herself for treatnt on September 28, 1901. She stated that she had undergone a newhat serious operation some 21 months previously. Since the date that operation she had suffered from a succession of very painful deepted boils (a reference to her diary showed that the exact number of ese had been 25). In addition she had constantly suffered from superial boils and pimples which had made sitting uncomfortable. She nplained also of a pustular discharge occurring intermittently from her se and ears. The patient was for the moment free from deep-seated ils. A sample of blood was withdrawn for examination and the patient s inoculated with 0·75 cubic centimetre of staphylococcus vaccine.[1]

Data of the Blood Examinations undertaken before Inoculation.— *timation of the inhibitory power exerted by the serum on the growth staphylococcus.*—For the purposes of this estimation a 24-hour old oth cultivation of the staphylococcus aureus was diluted [2] with liquefied trient gelatin until a dilution of 1 in 1,000,000 had been arrived at. a five-cubic-millimetre volume of this dilution was then added in each se an equal volume of (*a*) the patient's serum, (*b*) the patient's serum er this had been heated for ten minutes to 60° C., (*c*) serum from a normal rson, and (*d*) sterile nutrient broth. After these admixtures had been ide the diluted culture was in each case drawn up into a capillary tube. ree such tubes were being filled in with each variety of admixture. ble III shows the numbers of colonies [3] which developed.

Here manifestly the same results are obtained as in the case of the imations undertaken before inoculation in connexion with Patient 1 *de supra*). In other words, no indication was obtained of anything in e nature of an inhibitory effect being exerted upon the staphylococcus either the control blood or the patient's blood. We shall see in nnexion with the next cases that the difference between the blood of

[1] This vaccine was a suspension in normal salt solution of a 24-hour old culture the staphylococcus aureus subcultured from Patient 1. The suspension was rilized at 65° C. The quantum injected corresponded to the quantum of culture ich was obtained from 0·75 square centimetre of agar surface.

[2] The dilutions were made with the diluting pipette figured by me in *The Lancet* June 1, 1901, p. 1532.

[3] The colonies were counted under the microscope by the technique described *The Lancet* of December 1, 1900, p. 1556.

patients suffering from staphylococcus invasions and the blood of norm persons is to be sought elsewhere.

TABLE III.—*Showing the Number of Staphylococcus Colonies wh Developed in Capillary Tubes filled in with Five Cubic Millimet of the Diluted Gelatin Culture of Staphylococcus mixed with five Cu Millimetres of Serum or Broth, as Particularized in Column 1 bel*

Admixtures which were made to the 1,000,000-fold dilution of Staphylococcus Culture.	Number of Staphylococcus Colonies which developed in each capillary tube.			
	Tube 1.	Tube 2.	Tube 3.	Average three tub
Patient's serum	10	15	10	11·6
Patient's serum previously heated to 60° C. for 10 minutes . . .	9	6	20	11·6
Normal serum	11	13	11	11·6
Sterile broth	18	9	16	14·3

Particulars with regard to the subsequent inoculations and resulti clinical symptoms.—The first inoculation was followed by very little the way of either local or constitutional symptoms. Two further inoc lations, in each case with one cubic centimetre of the same vaccine, w undertaken within the next three weeks. As in the case of the fi inoculation the symptoms were very slight.

Data of Blood Examinations undertaken a Week after the Third Inoc lation. *Estimation of the agglutination power exerted by the blood upon staphylococcus.*—The patient's blood now gives a complete sedimentati in an eight-fold dilution. The results obtained with the control blo are, however, indefinite.

Estimation of the bactericidal power.—A 1,000,000-fold dilution o 24-hour old broth culture of the staphylococcus having been made, measured volumes of this were transferred to the surface of agar. the first of the six agar tubes (inoculated with 25 cubic millimetres of diluted culture) nine colonies of staphylococcus made their appearan In the second and third tubes (inoculated in each case with 15 cu millimetres) the number of colonies which developed were in each case s In the fourth tube (inoculated with 10 cubic millimetres) two colon made their appearance. The fifth and sixth tubes (inoculated in ea case with five cubic millimetres) remained sterile. This gives an avera of 3·07 colonies to each 10 cubic millimetres of the 1 in 1,000,000 diluti employed. Figuring this out we arrive at 307,000,000 as the number staphylococci contained in a cubic centimetre of the undiluted culture.

The procedures connected with the enumeration of the culture havi been completed, graduated dilutions of the original staphylococcus cultu were made and were mixed in capillary tubes with the patient's serum a a normal serum respectively. In each case about five cubic millimet of serum and diluted culture were employed. After an interval of

s, during which the tubes were digested at 37° C., the capillary tubes filled in[1] with sterile broth with a view of determining whether any ericidal effect had been exerted. The results are subjoined (Table IV). t will be manifest from these results set forth above that neither the nal serum nor the patient's serum exerted any bactericidal action n the growth of the staphylococcus. Comparison with the result of enumeration shows that five cubic millimetres of either serum failed ill 1·5 staphylococci.

LE IV.—*Showing the Absence of a Bactericidal Power in the Patient's Blood after these Inoculations.*

Dilutions of the Staphylococcus Culture which were digested with the Serum.	Results obtained on incubating the Tubes after filling in with Sterile Broth.	
	Tubes containing the Patient's Serum.	Tubes containing Normal Serum.
100-fold dilution .	Growth of Staphylococcus	Growth of Staphylococcus
1,000-fold " .	" "	" "
10,000-fold " .	" "	" "
00,000-fold " .	" "	" "
00,000-fold " .	" "	" "
00,000-fold " .	Tube remains sterile	Tube accidentally contaminated

Particulars with regard to the patient's condition after inocula- n and estimations of the phagocytic power of the blood.—The patient orded that she felt herself better within 24 hours after the first inocula- n. For the first time for months she could sit with ease. There was a complete cessation of the purulent discharge from the nose and the . The general health also is said to have markedly improved and the ient was able to go about and to enjoy life. Two months from the e of the first inoculation the patient presented herself for examination. had not had any recurrence of the boils, but had been troubled by a irritable pimples on the hands. A sample of blood having been hdrawn from the finger an estimation of the phagocytic power of the od was undertaken. The results of the estimation are subjoined ble V).

In view of the result of this estimation the patient was warned that ecurrence of the boils must probably be anticipated. After a free erval of three and a half months the patient, early in January, 1902, eloped a deep-seated boil. This last ran a similar course and was ended by as much pain as the deep-seated boils which had occurred ore the date of inoculation. The patient was seen again on Janu- 28, 1902. Her condition was then quite satisfactory, but she still fered from occasional irritable pimples. An estimation of the phagocy-

[1] The technique that was employed was that described by me in *The Lancet* June 1, 1901, p. 1532.

tic power of the blood carried out at room temperature gave the followi results : average number of staphylococci ingested by a white blood co puscle of a normal blood,[1] 4·6 ; average number ingested by a whi blood corpuscle of the patient's blood, 3·5. The proportion of staphyl cocci ingested by normal blood to staphylococci ingested by patien blood was thus 1 : 0·75.

On March 17 the patient reported that she had no recurrence of t boils and that she was no longer troubled with pimples.

TABLE V.—*Showing the Phagocytic Power of Patient Two Months af Inoculation.*

Variety of Staphylococcus which was employed.	Average Number of Staphylococci ingested by each Polynuclear White Blood Corpuscle of the *Control* blood.	Average number of Staphylococci ingested by each Polynuclear White Blood Corpuscle of the *Patient's* Blood.	Phagocytic Index—i.e., th Proportion in which the Number of Staphylococci i gested by *each Polynuclea White Blood Corpuscle of Control* blood stands to th Number of Staphylococci ingested by the Polynucle White Blood Corpuscles of *Patient's* Blood.
Staphylococcus aureus subcultured from Patient 1	25·7	10·6	1 : 0·41
Staphylococcus albus subcultured from Patient 3	39·3	21·6	1 : 0·55

CLINICAL AND BACTERIOLOGICAL DATA RELATING TO CASE 3.

The patient, a medical man, about 30 years of ago, presen himself for treatment on December 6, 1901. He had suffered contin ously for the previous 12 months from small but very irritable pustu pimples and superficial boils on the back of the neck. He was still suff ing in this way. A cultivation made from one of his boils yielded a pu cultivation of staphylococcus albus.

Data of Blood Examinations instituted before Inoculation.—A cou of the different varieties of white blood corpuscles showed that t relative numbers were as follows : Eosinophil, 1 per cent. ; polyn clear, 53 per cent. ; basophile, 1 per cent. ; and large mononuclear a lymphocytes, 45 per cent. In an estimation of the phagocytic power the blood the results shown in Table VI were obtained.

It will be seen that the phagocytic power of the patient's white blo corpuscles was between two and three times less than the phagocy power of the white blood corpuscles of a normal blood.

Particulars of the inoculations which were carried out and the clini symptoms resulting from these.—On November 11, 1901, the patient w

[1] This blood was derived from A. E. W.

ılated with one cubic centimetre of the same staphylococcus vaccine h was employed in Case 2 (*supra*). The symptoms at the site of ılation were comparatively trifling; the patient, however, felt a out of sorts for two days. On December 1 the patient was inoculated 0·75 cubic centimetre of a staphylococcus vaccine prepared from cultures of the staphylococcus albus subcultured from himself. This tum of vaccine represented the quantum of culture which developed ·5 square centimetres of agar surface. Marked local reaction was ıced at the site of inoculation. The constitutional reaction was only slight.

ᴇ VI.—*Estimation of the Phagocytic Power of the Blood of the Patient in Case 3 anterior to Inoculation.*

—	Average Number of Staphylococci ingested by each Polynuclear White Blood Corpuscle of the *Normal* Blood.	Average number of Staphylococci ingested by each Polynuclear White Blood Corpuscle of the *Patient's* Blood.	Phagocytic Index—i.e., the Proportion in which the Number of Staphylococci ingested by each White Blood Corpuscle of *Normal* Blood stood to the Number of Staphylococci ingested by each White Blood Corpuscle of the *Patient's* Blood.
estimation .	16·6	6·5	1 : 0·39
d estimation.	21·0	8·0	1 : 0·38

articulars with regard to the effects produced upon the blood and upon ocalized staphylococcus invasion.—The effects exerted upon the condi- of the neck and the concurrent changes in the blood are set forth v with some detail in parallel columns (Table VII). The results of blood examination are also presented in a synoptical manner in the of a graphic curve (*Chart* 1).

hree important points are clearly brought out by the observations e upon the patient. (1) The first of these points is that changes in phagocytic power of the blood in the form of a negative phase of nished phagocytic power, succeeded by a positive phase of increased ocytic power follow upon a staphylococcus inoculation. The tive and positive phases here obtained are plainly precisely analogous e negative and positive phases of bactericidal power which I have n to supervene upon an anti-typhoid inoculation. They are analo- also to the negative and positive phases which Ehrlich and Madsen shown to supervene upon the inoculation of tetanus and diphtheria respectively. (2) The second and equally important point which is ght out by the comparison of the clinical record with the results of blood examination is that the negative and positive phases of phagocy- ower stand in the very closest relation to the resisting power of the nism. So close and intimate does that association appear to be that s, in this and the subsequent cases, always easy from the consideration

TABLE VII.—*Setting forth in connexion with Case* 3 (a) *the Phagocytic Power of the Patient's Blood*; (b) *the Details of the Treatment adopted*; *and* (c) *Particulars with regard to the Patient's Clinical Condition.*

Date.	Variety of Staphylococcus employed in Testing the Blood.	Phagocytic Index—i.e. the proportion in which the Number of Staphylococci ingested by each Polynuclear Leucocyte of the *Normal* Blood stood to the Number of Staphylococci ingested by each of the *Patient's* Polynuclear Leucocytes.	Brief Notes regarding the Localized Staphylococcus Invasion and the General Condition of the Patient.
1901. Nov. 9	Staphylococcus aureus from the patient in Case 1	1* : 0·39	For description of clinical condition *vide* text (*supra*).
	Inoculated in the flank with 1 c.c. of staphylococcus vaccine prepared with staphylococcus aureus subcultured from the patient in Case 1.		
Nov. 10	Staphylococcus aureus from the patient in Case 1	1† : 0·92	Very slight local or constitutional disturbance. Condition of the neck unaltered.
„ 11	„ „ „ „ „	1† : 0·3	Marked improvement in the pustular pimples on the neck.
„ 14	„ „ „ „ „	1† : 0·6	The boils practically gone. The patient felt better.
„ 15	„ „ „ „ „	1† : 1·35	No return of the boils.
„ 16	„ „ „ „ „	1† : 1·95	„ „ „
„ 17	„ „ „ „ „	1‡ : 1·25	„ „ „
„ 18	„ „ „ „ „	1† : 1·45	„ „ „
„ 20	„ „ „ „ „	1† : 0·88	A fresh development of small boils on the neck.
„ 21	„ „ „ „ „	1 : 0·85	The boils are drying up.
„ 22	„ „ „ „ „	1 : 2·5	One small fresh boil; the others improving.
	Staphylococcus albus from the patient in Case 3	1 : 1·3[illegible]	
„ 23	Staphylococcus aureus „ „ „ 1	1 : 1·6	No new boils, all the old ones have dried up.
	Staphylococcus albus „ „ „ 3	1 : 0·9	
„ 25	Staphylococcus aureus „ „ „ 1	1 : 1·1	No return of the boils.
	Staphylococcus albus „ „ „ 3	1 : 0·5[illegible]	
„ 26	Staphylococcus aureus „ „ „ 1	1 : 1·3	No new boils, but some irritation round one of the old ones.
	Staphylococcus albus „ „ „ 3	1 : 1·6	
„ 27	Staphylococcus aureus „ „ „ 1	1 : 1·0[illegible]	No fresh boils.
	Staphylococcus albus „ „ „ 3	1 : 1	
„ 30	Staphylococcus aureus „ „ „ 1	1 : 1·5	No fresh boils, but there is still some infiltration of the neck.
	Staphylococcus albus „ „ „ 3	1 : 1	
Dec. 1	Staphylococcus aureus „ „ „ 1	1 : 1·5	A small boil has developed on the right side of the neck.
	Staphylococcus albus „ „ „ 3	1 : 0·[illegible]7	

Inoculated in the flank with 0·75 cubic centimetre of a vaccine prepared from his own staphylococcus albus. The quantity inoculated represents the quantum of culture obtained from 1·5 square centimetres of agar surface.

Dec. 2	Staphylococcus aureus from the patient in Case 1	1 : 0·67	Marked local reaction at the site of inoculation. Slight constitutional disturbance.
	Staphylococcus albus " " " 3	1 : 0·75	
" 3	Staphylococcus aureus " " " 1	1 : 0·94	Local reaction at site of inoculation subsiding.
	Staphylococcus albus " " " 3	1 : 0·66	
" 4	Staphylococcus aureus " " " 1	1 : 0·53	Still some pain at site of inoculation. A painful boil has developed on the neck. This is the first painful boil since the patient came under treatment.
	Staphylococcus albus " " " 3	1 : 0·47	
" 6	Staphylococcus aureus " " " 1	1 : 1·65	The boil which developed on Dec. 4 is much better.
	Staphylococcus albus " " " 3	1 : 1·1	
" 8	Staphylococcus aureus " " " 1	1 : 1·52	Two or three small but somewhat pustular pimples have developed.
	Staphylococcus albus " " " 3	1 : 1	
" 9	Staphylococcus aureus " " " 1	1 : 1·1	A few fresh small pimples on the neck.
	Staphylococcus albus " " " 3	1 : 1·15	
" 15	Staphylococcus aureus " " " 1	1 : 1·05	The patient has been free from boils for some days past. Infiltration of the neck has been steadily diminishing.
	Staphylococcus albus " " " 3	1 : 1·05	
" 18	Staphylococcus aureus " " " 1	1 : 0·54	Some fresh pimples have developed.
	Staphylococcus albus " " " 3	1 : 1·1	
" 27	Staphylococcus aureus " " " 4	1§ : 3·9	All infiltration has disappeared. Except for the presence of slight congestion the skin is absolutely normal.
1902. Jan. 3	Staphylococcus albus " " " 3	1 : 1·2	The patient has been free from boils for some little time. His neck is perfectly smooth and clean. The patient is now proceeding to South Africa.

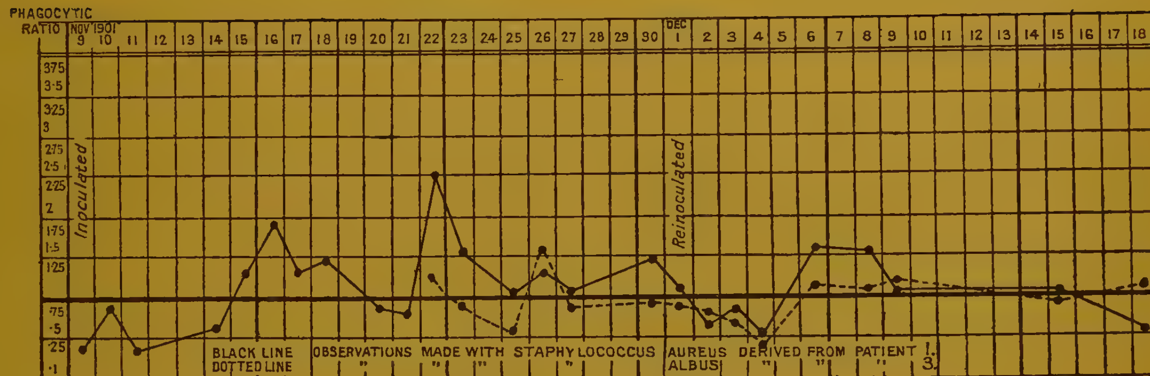
* In this and all subsequent estimation, unless where otherwise specified, the control blood employed was derived from W.B.L.

† The control blood used in this estimation was derived from F. N. W.

‡ Three normal bloods, those of W. B. L., F. N. W., and X. A., were here employed as controls. The results obtained in the case of these bloods were in the closest agreement.

§ The blood here employed was derived from A. E. W. It is a blood which possesses a low phagocytic power.

Chart 1 (relating to Patient 3).



The curves express the phagocytic indices of the patient's blood (as estimated by Leishman's method), the phagocytic power of the control blood being everywhere taken as 1.

TABLE VIII.—*Setting forth in Connexion with Case 4 (a) the Phago[illegible] the Treatment adopted; and (c) Particulars with regard to the Clinical Condition of the Patient.*

Date.	Variety of Staphylococcus employed in the Test.	Phagocytic Index—*i.e.* the Proportion which the Number of Staphylococci ingested by White Blood corpuscles of *Normal* Blood bore to Number of Staphylococci ingested by *Patient's* Blood.	Brief Notes regarding the Conditions of the Localized Staphylococcus Invasion and the General Condition of the Patient.
1901. Dec. 20	Staphylococcus albus from the patient in Case 3	1* : 0·78	Condition as described in the text (*supra*).
Inoculated with 0·3 cubic centimetre of a staphylococcus vaccine prepared from agar cultures of staphylococcus albus subcultured from the patient in Case 3. The quantum inoculated corresponds to the quantum of culture which developed on 0·6 square centimetre of agar surface.			
Dec. 21	Staphylococcus albus from the patient in Case 3	1 : 2	There has been very little local reaction at the site of inoculation and only slight constitutional disturbance. Considerable swelling has, however, developed in the neck. Signs of improvement in the fact that the superficial boils have lost their angry look. There is considerable polynuclear leucocytosis.
	Staphylococcus aureus derived from the patient himself	1 : 3·8	
,, 24	Staphylococcus aureus from the patient himself	1† : 6·48	The patient reports that there has been very considerable inflammatory swelling of the neck and of the lymphatic glands. The swelling has now almost subsided. The boils have entirely disappeared and the leucocytosis has passed off.
Inoculated with 0·5 cubic centimetre of a vaccine prepared from agar culture of the patient's own staphylococcus aureus. The quantum inoculated corresponds to the quantum of culture which developed in 0·5 square centimetre of agar surface.			
Dec. 27	Staphylococcus aureus from the patient himself	1 : 2	No constitutional reaction. Congestion and tenderness for 48 hours at the seat of inoculation. No further swelling of the glands or inflammatory reaction in the neck. The patient is quite free from boils and acne. There is some leucocytosis.
1902. Jan. 8	,, ,, ,, ,,	1 : 1	One small inflamed pimple has developed on the neck. The patient otherwise is quite well, and there is no leucocytosis.
,, 10	Staphylococcus albus from the patient in Case 3	1‡ : 0·95	The neck is perfectly well. No trace remains of boils, acne, or swollen glands.
,, 17	—	—	The patient, who is leaving the neighbourhood, reports himself as being perfectly well. A similar report was received on March 15, 1902.

* The control blood employed in this and the five following estimations was derived from A. E. W.

† This result may conceivably have been due to some error of technique or to some incidental lowering of the phagocytic power of the control blood. *Vide* in this connexion the remarks appended to Table IX and Curve 2.

‡ The control blood here employed was derived from W. B. L. The phacocytic power of this blood has invariably been found to be higher than that of A. E. W.'s blood.

connexion with Case 5 (a) *the Phagocytic Power of the Blood;* (b) *Details with regard*
nt adopted; and (c) *Particulars with regard to the Clinical Condition.*

	...ococcus employed in testing the Blood.	Phagocytic Index—i.e., Proportion which the Number of Staphylococci ingested by the white blood Corpuscles of the *Normal* blood bore to the number of Staphylococci ingested by the White Corpuscles of the *Patient's* Blood.	Brief Notes regarding the Condition of the Localized Staphylococcus Invasion and the General Condition of the Patient.
	Staphylococcus aureus from the patient in Case 4	1* : 3·3	For description of condition see text (*supra*).
...oculated in the flank with 0·5 cubic centimetre of vaccine prepared from agar cultivations of staphylococcus aureus subcultured from the patient in Case 4. The culture inoculated corresponded to the amount of culture which developed in 0·5 square centimetre of agar surface.			
Dec. 25	Staphylococcus aureus from the patient in Case 4	1* : 0·16	Great swelling and inflammatory congestion over the whole affected area of the face, the neck, and the scalp. Considerable leucocytosis.
„ 26	„ „ „ „ „	1* : 0·34	Still some swelling. A sensible decrease in the pustular points and on the cheeks and the chin.
„ 27	„ „ „ „ „	1* : 2·7	The inflammatory reaction has subsided. All the pustular points have disappeared. Polynuclears 50 per cent.
„ 28	„ „ „ „ „	1* : 1·3	The face is much better. The eyelids are no longer red. No further development of the pustular points.
Inoculated in the flank with 0·5 cubic centimetre of the same staphylococcus vaccine.			
„ 29	Staphylococcus aureus from the patient in Case 4	1* : 0·88	Renewed swelling and congestion of the face. A few pustular points on the chin. Marked improvement everywhere.
„ 30	„ „ „ „ „	1* : 1·94	Congestion less. Four or five pustular points on the chin.
1902. Jan. 2	„ „ „ „ „	1* : 0·94	Complaints of more soreness and swelling. The face more congested, and a new crop of pustular points and more serous weeping.
„ 3	Staphylococcus albus from the patient in Case 3	1* : 1·0	No change noted except a slight diminution in the swelling.
„ 6	„ „ „ „ „	1† : 0·55	The chin is very angry looking; more pustular points and weeping.
„ 7	„ „ „ „ „	1† : 0·61	The face is less angry.

Inoculated with 0·5 cubic centimetre of vaccine prepared from agar cultivations of the patient's staphylococcus aureus.

,, 8	Staphylococcus albus from the patient in Case 3	1† : 1·66	Considerable inflammatory congestion of the chin, subparotid region, eyelids and temples. The general appearance is almost erysipelatous.
,, 9	,, ,, ,, ,, ,,	1* : 2·0	Swelling and infiltration much diminished. The bony contours of the chin and the angle of the jaw have become clearly visible.
,, 10	,, ,, ,, ,, ,,	1† : 0·86	The face and the chin are rather more inflamed. Had a bad night. Swelling round the glands below the angle of the chin.
,, 11	,, ,, ,, ,, ,,	1† : 1·74	Marked improvement everywhere. The swathings and moist boric dressings are now discarded, and the face is powdered with chalk.
,, 12	,, ,, ,, ,, ,,	1† : 1·46	Infiltration everywhere much diminished.
,, 14	,, ,, ,, ,, ,,	1† : 1·55	No change. The crusts which still cover the chin are removed by steeping in normal salt solution.
,, 16	,, ,, ,, ,, ,,	1† : 0·8	Increased congestion with weeping and scabbing. No pustulation.
		Inoculated with one cubic centimetre of the same staphylococcus vaccine.	
,, 17	Staphylococcus albus from the patient in Case 3	1† : 1·35	A red blush has appeared over the whole of the affected area. There is no noticeable swelling. The chin is thickly encrusted with scabs.
,, 18	,, ,, ,, ,, ,,	1† : 1·73	No change. The crusts were removed by steeping in normal salt solution.
,, 19	,, ,, ,, ,, ,,	1† : 1·18	Great improvement; appearance of healthy cicatrization over the area of the chin.
,, 20	,, ,, ,, ,, ,,	1† : 1·6	The cheeks are better, but there is an increased serous discharge on the skin.
		Inoculated with two cubic centimetres of the same staphylococcus vaccine.	
,, 21	Staphylococcus albus from the patient in Case 3	1† : 0·6	Inflammatory reaction, swelling, and increased serous exudation. These symptoms are confined to the region of the chin.
,, 22	,, ,, ,, ,, ,,	1† : 1·3	The face and the chin are very much better. Infiltration has largely disappeared, and the skin looks delicate and soft. It is of a pink colour over the whole of the previously affected area. A single small gland can be felt under the angle of the left jaw. There is still a little weeping over the surface of the chin, and there are some crusts in the scalp above and behind the ear.
,, 23	,, ,, ,, ,, ,,	1† : 1·3	The improvement is maintained.
,, 24	,, ,, ,, ,, ,,	1† : 1·3	The surface of the chin is becoming normal.
,, 25	A mixture of staphylococcus aureus and albus	1† : 1·85	The patient is improving rapidly, the skin is everywhere soft and pliable and is less flushed.
,, 30	,, ,, ,, ,, ,,	1† : 0·61	About ten new superficial pustular points have appeared on the chin; the pustular eruption over the temporal area is drying off.

Inoculated with three cubic centimetres of staphylococcus vaccine.

* The control blood employed in these estimations was derived from A. E. W.
† The control blood employed in these estimations was derived from W. B. L. It is a blood which possesses a high phagocytic power. *Vide* in connexion with results of phagocytic estimation on December 24, 1901, note † appended to Table VIII.

TABLE X.—*Setting forth in Connexion with Case* 6 (a) *the Phagocytic Power of the Patient's Blood* ; (b) *the Details of the Treatment adopted* ; *and* (c) *Particulars with regard to the Patient's Clinical Condition.*

Date.	Variety of Staphylococcus employed in testing the blood.	Phagocytic Index—i.e. the Proportion which the Number of Staphylococci ingested by the Leucocytes of the *Control* Blood bore to the Number of Staphylococci ingested by Leucocytes of the *Patient's* Blood.	Brief Notes regarding the Conditions of the Localized Staphylococcus Invasion and the General Condition of the Patient.
1902. Feb. 18	Staphylococcus albus subcultured from the patient in Case 3	1* : 0·74	Condition as described in the text (*supra*).
„ 19	Staphylococcus albus subcultured from the patient in Case 3	1* : 0·7	Condition unchanged.
Inoculated in the flank with one cubic centimetre of a mixed staphylococcus vaccine. The quantum inoculated corresponds to the quantum of albus and aureus culture which developed on one square centimetre of agar surface.			
Feb. 20	Staphylococcus albus as above	1* : 1·24	The patient had a mild general and local reaction. The condition of the chin and the cheeks is greatly improved, there being much less congestion and a small amount of discharge.
„ 21	—	—	The condition of the chin, the face, and the scalp is very considerably improved.
„ 22	Staphylococcus albus as above.	1† : 0·87	Further marked improvement, the back of the head and of the chin having become practically clear. Small moist patches still to be seen in front of the ear and at the angle of the mouth.
Re-inoculated with one cubic centimetre of the same vaccine.			
„ 23	Staphylococcus albus as before	1† : 0·36	The moist patch in front of the ear has dried up. The patient is quite well with the exception of a small patch at the angle of the mouth.
„ 24	„ „ „	1† : 0·36	Practically well.
„ 25	„ „ „	1* : 0·7	Discharged.

* The control blood employed was that of W. B. L.
† The control blood employed was that of W. G. L.

he local appearances to anticipate the results of the phagocytic nation. (3) The general result of the blood examinations is in entire rdance with the theory of immunity put forward by Metchnikoff.

CLINICAL AND BACTERIOLOGICAL DATA RELATING TO CASE 4.

The patient, aged 21 years, a medical student, presented himself for tment on December 20, 1901. For the previous two months he had suffering from boils, chiefly on the nape of the neck, but also on the lder and forearm. Immediately anterior to the commencement of boils the patient had suffered from an attack of influenza. The of the neck was seen to be the seat of a localized inflammatory process, surface being occupied by three or four angry pimply boils, while deeper tissues were considerably infiltrated and tumefied. The glands oth sides of the neck were somewhat swollen. On the cheek, in the hbourhood of the nose, there were a certain number of very prominent s of pustular acne. A cultivation made from one of the pustular ples on the back of the neck yielded a pure cultivation of the staphy-ccus aureus.

Particulars of the inoculations undertaken, of the resulting symptoms and d changes, and of the effect produced on the localized staphylococcus sion.—The course of treatment pursued and the effects which resulted that treatment will perhaps be best gathered from a consideration e subjoined tabular statement (Table VIII).

The general deductions which can be drawn from a consideration of case are precisely the same as those drawn from a consideration of 3. The absence of a negative phase in this particular case is no doubt e imputed to the fact that very small doses of staphylococcus vaccine employed. Interesting as bearing on the association between changes in the phagocytic power of the blood and the changes in the dition of the patient are (1) the rapidity with which the therapeutic lt was achieved taken in connexion with the rapid increase in the gocytic power of the blood, and (2) the coincidence of the development e pustular pimple on January 3 with the result of the blood examina- instituted on that day. A further and very interesting feature which sts attention in connexion with this case is the fact that the first ulation was followed by considerable inflammatory swelling at the of infection. This phenomenon, which, as we shall see, made its earance again in a very marked manner in connexion with Case 5, is henomenon which would seem to be precisely analogous to the local ammatory reaction which is obtained after the injection of tubercle cine (Koch's tuberculin). It will be noticed in this connexion that no ammatory reaction supervened upon the second inoculation, and erally that nothing of this kind has come under notice in connexion h inoculations, undertaken upon patients whose tissues were pre-

sumably at the date of inoculation free, or comparatively free, fr staphylococcus.

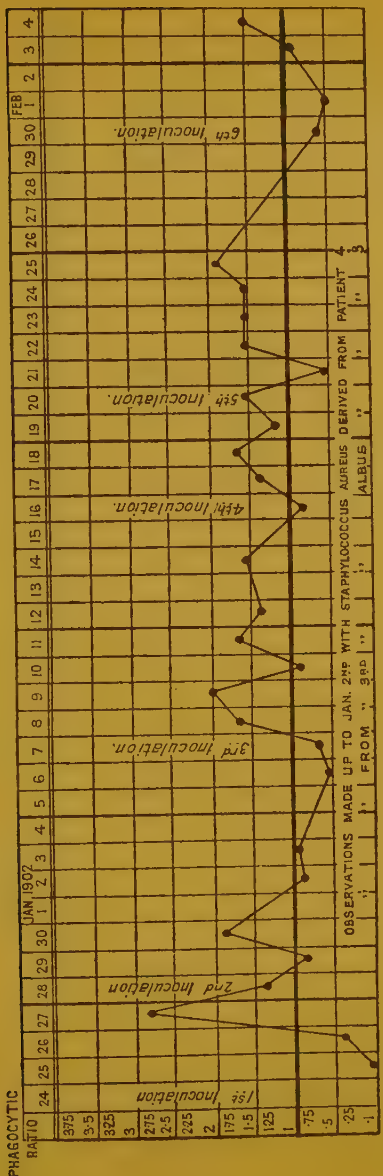
CLINICAL AND BACTERIOLOGICAL DATA RELATING TO CASE 5.

The patient, a private soldier, invalided home from Ra Pindi for intractable sycosis, was first seen on December 24, 1901. gave the following history. In 1893 a little sore developed on scalp and this was followed by a general eruption all over the scalp a face. He was admitted to, and remained under treatment in, the Camb well Infirmary for six weeks and was then discharged "cured." When enlisted in 1897 he had, however, still a minute patch of eruption in fro of his left ear. After serving for six months he was admitted to hospi for a deep-seated boil in the left parotid region which developed in co nexion with the uncured patch of eruption just spoken of. He remain under treatment for about a fortnight. In the same year he proceed to India and remained well for a time. In 1899 he attended hospi for three months for sycosis and eczema of the face. He relapsed aga in 1900 and spent nine weeks in hospital, afterwards attending as an ou patient. He was readmitted in 1901, and was invalided home after spen ing six or seven months in hospital under energetic treatment. T patient, whose face and head were entirely swathed in dressings, w suffering from a very aggravated form of sycosis complicated by eczem The whole area of the chin, the cheeks, and the under surface of the ja was extensively infiltrated and was thickly covered in places with moi scabs. A number of pustular points were interspersed between t prominences corresponding to the inflamed hair follicles. The bo contours of the jaw were almost obliterated by dense masses of infiltrat tissue. The scalp and in particular that portion of it overlying t squamous portion of the left temporal bone was the seat of a scabb sero-pustular affection corresponding to that described under the nam of acne varioliformis. The eyebrows and the margins of the eyeli were the seat of a chronic pustular inflammation.

Data of the microscopical and bacteriological examinations which we instituted.—A careful examination of the hairs of the beard showed th these were not invaded by any parasitic mycelium. Cultivations mad from the pustular points on the face yielded pure cultivations of the staph lococcus aureus.

Details of the inoculations undertaken and of the effects of these upon th blood and the localized staphylococcus invasion.—The particulars of th inoculations undertaken and of the effects produced on the blood and o the area invaded by the staphylococcus are subjoined with some deta in tabular form (Table IX). The results of the blood examinations ar given also in the form of a curve (*Chart* 2).

CHART 2 (relating to Patient 5).



The curve expresses the phagocytic index of the patient's blood, the phagocytic power of the control blood being everywhere taken as 1.

Attention may be drawn to the following points in connexion with the table and the curve. (1) The high expression obtained for the phago-

cytic power of the patient's blood in the case of the first estimation recorded in the table would seem to have been due either to an underestimation of the phagocytic power of the control blood due to an error in the technique or to some incidental lowering of the phagocytic power of that blood. There are thus in this case no trustworthy data relating to the phagocytic power of the patient's blood before inoculation. (2) It will be observed that, as in the cases already considered, an increase of phagocytic power was registered after each inoculation. (3) In the case of the first and second and again in the case of the fifth and sixth inoculations undertaken respectively with a double and a treble dose of the vaccine, the positive phase was preceded by a negative phase of diminished phagocytic power. (4) The clinical record shows that a condition of diminished phagocytic power, whether produced by inoculation or occurring spontaneously, was in each case associated with aggravation of the local affection. (5) The clinical record further shows that a condition of increased phagocytic power was invariably associated with an inflammatory (phagocytic) reaction in the area invaded by the staphylococcus and with a subsequent improvement in the form of a restriction of the invaded area. (6) It would appear from the consideration of the present case, taken in association with the cases already considered, that where the scale is turned against the invading staphylococci, it is turned not by a change produced in the blood-fluids, but by a succession of immunising impulses, each such impulse expressing itself in an inflammatory (phagocytic) reaction localized in the invaded tissues.

The results of the continuation of the treatment up to March 15 have confirmed the premonitions of a proneness to relapse which may be gathered from the previous history as well as from the results of the blood examinations and clinical data incorporated in the table and curve. In each case, after an improvement extending over three or four days subsequently to inoculation, symptoms of a certain amount of relapse have manifested themselves both in the condition of the local infection and in the results of the blood examinations. At present, after ten further staphylococcus inoculations which have been timed, and varied with respect to dose, in accordance with the results of the daily blood examinations, the patient's conditions is still that of a chronic staphylococcus invasion associated in places with a certain amount of scabbing and infiltration. There would thus appear to be a very definite limit to the defensive reaction of the organism, a limit which comes into consideration also in the case of other inoculations.

CLINICAL AND BACTERIOLOGICAL DATA RELATING TO CASE 6.

The patient, a man about 40 years of age, was first seen on February 18, 1902. A fortnight previously he began to suffer from an acute inflammation of the hair follicles, associated with a considerable

ount of serous discharge and itching. The inflammatory process v occupies the whole area of the beard and hairy portion of the cheeks, l it has spread backwards and has affected the nape of the neck and the ver portion of the back of the scalp. The whole of the affected area thickly occupied by discrete, for the most part dry, scabs conforming the type of acne varioliformis. Associated with these there is in places ertain amount of serous discharge. The scalp generally is extremely rfy. Cultivations made from the surfaces exposed by removal of the bs yielded a pure cultivation of staphylococcusaureus. The particus of the treatment and details as to the result obtained are subjoined Table X.

It will be noted on study of the above table that there was a marked rease in the phagocytic power of the patient's blood on the day sucding the inoculation. On this and the next two days the clinical ndition improved in an astonishing manner. The phenomena which pervened upon reinoculation are of less conspicuous interest. It will be n that after the second inoculation, which was resorted to with a view checking the falling away of the phagocytic power which manifested elf on the third day after the first inoculation, a pronounced negative ase of diminished phagocytic power came under observation. This gative phase, presumably owing to the fact that the invading bacteria d already been disposed of, was unaccompanied by anything in the ture of an aggravation of the patient's symptoms. At the date of the tient's discharge the negative phase was apparently giving place to a sitive phase of increased phagocytic power.

nsiderations in Connexion with the Possible Prophylactic and Therapeutic Applications of Anti-Staphylococcus Vaccine.

Having in the foregoing considered the results obtained by the inocuion of staphylococcus vaccine into patients affected with furunculosis, cosis, and acne, we may naturally be led on to inquire whether there are y other forms of staphylococcus invasion in connexion with which ese inoculations might perhaps find a useful application.

Possible prophylactic applications.—Dealing first with the question of possible application of these vaccines in connexion with the prophylaxis septic invasion, it would seem that advantage might possibly be derived om their application in connexion with compound fractures, and further connexion with operative procedures, such as those associated with the per jaw and air passages, where it is impossible to secure the asepticity the wound. It is, perhaps, venturesome to suggest that there might so be other cases where the surgeon might be willing in his operative ocedures to avail himself of the increased resistance to staphylococcus vasion which would, it can hardly be doubted, be obtained by a prophyctic inoculation of staphylococcus vaccine. I would urge in defence of is suggestion that the adoption of measures designed to increase the

resistance of the organism to septic invasion would seem to be only a legit mate development of the accepted practice of relying upon the protectiv agencies of the organism instead of upon the application of antisepti for the destruction of such septic bacteria as may upon occasion in spite aseptic precautions obtain access to wounds.

Apart from actual surgical procedures it would seem possible th prophylactic inoculation of staphylococcus vaccine might find a scient fically interesting and conceivably a practically useful application connexion with anti-small-pox vaccination. In view of the apparent septic character of the inflammation which is frequently associated wi vaccination wounds, and the fact that staphylococci are constant present in vaccine lymph, even in glycerinized lymph, I take it that would be desirable to determine how far the staphylococcus is responsib for the symptoms which have done so much to discredit vaccination the popular mind. The information which is required could probab be obtained by noting the effects produced by the inoculation of one a the same variety of vaccine lymph into persons or animals immunise against the staphylococcus and into persons or animals not so pr tected.

Comparatively little remains to be added to what has already co under consideration in connexion with the therapeutic employment staphylococcus vaccine. It is possible that the vaccine might find a us ful application, not only in connexion with the treatment of furunculos acne, and sycosis, but also in connexion with the treatment of Vel sores, old ulcers, sinuses, and septic vaccination wounds.

Concluding Remarks.

Possibility of a still wider extension of the treatment of localized infla matory conditions by inoculation.—We may inquire, in conclusion, in the prospects of successfully exploiting bacterial vaccines in the treatme of localized inflammatory processes produced by pathogenic micr organisms other than the staphylococcus. Much that is of value can learned upon this subject by a careful consideration of Koch's tubercul inoculations and by a comparison of these with the inoculations of staph lococcus vaccine with which we have been dealing above. In each case bacterial vaccine is, for therapeutic purposes, inoculated into patien already the subject of a corresponding infection. In each case, as t result of the inoculation, an acute inflammatory reaction is set up at t seat of infection. In each case, again, as a result of the inflammato reaction in question, the nidus in which the bacteria are lodged is brok up.

At this point an all-important difference emerges. In the case whe the localized inflammation process is due to a staphylococcus infectio

e breaking up of the nidus has involved, so far as has hitherto appeared, destruction of the bacteria which have been fluttered. In the case of tubercle infection the destruction of the nidus has, on the other hand, en shown to be compatible with the continued survival of the tubercle cilli. Judging by the event recorded in connexion with many tuberculin oculations, the tubercle bacilli when set free by the inflammatory retion would appear to have been merely carried away by the white ood corpuscles or the lymph current to give origin to new localized ci of infection, or, in exceptional cases, to a generalized form of berculosis.

When we consider what may be the reason of this difference in the ent which has followed the inoculation of these two different bacterial ccines, we realize that it must be attributed primarily to the fact that e tubercle bacillus possesses, as compared with the staphylococcus, an initely greater capacity for maintaining its vitality in the interior of e organism.

A further reason for the difference in event we may perhaps find in the ct that while it was possible in the case of the staphylococcus inoculations scribed above to graduate the doses and time the injections in accordce with the data supplied by the measurement of the patient's resistce (as judged by his phagocytic reaction and by the condition of the vaded tissues) it was impracticable to do this—although it was infinitely ore important to do so—in the case of Koch's inoculations of tubercle ccine. We are thus by a comparison of the inoculations undertaken th staphylococcus vaccine with those undertaken with Koch's tubercu-, again led back to the cardinal principle that we must, in connexion th every therapeutic application of a bacterial vaccine, consider, on e one hand, the capacity of resistance with which the particular species invading micro-organism is endowed, and, on the other hand, the pacity of resistance possessed by the particular patient at the time of oculation.

The ideal to be kept in view must everywhere be so to graduate the ses of the bacterial vaccine and so to time the injections as to leave to e infected patient in any negative phase which may supervene after oculation a sufficient margin of resistance to safeguard him against any neralization of his infection.

In connexion with the possibility of a wider extension of the principle therapeutic inoculation we may with advantage keep in view (1) e consideration that patients who have been suffering from long-conued localized inflammation processes would seem, so far as can be judged m clinical experience, to have acquired a defensive power such as fices to ward off the more generalized forms of their particular infections, d (2) the consideration that where bacterial invasions manifest themves only under the form of " surface-invasions " the conditions in the erior of the organism must be assumed to be *ab initio* hostile to the owth of the invading micro-organisms.

These considerations would seem to point to the desirability of testi the effect of therapeutic inoculations of sterilized streptococcus cultu in the case of patients who are the subject of indolent and relapsi forms of erysipelas. In applying an inoculation treatment to the for of erysipelas referred to it would, of course, be necessary to keep in vi the fact that the streptococcus possesses, as compared with the staph lococcus, a much greater capacity for generalizing itself in the organism.

The above considerations would also seem to point to the desirabili of determining whether any therapeutic advantage could be deriv from the inoculation of the appropriate bacterial vaccines into patie suffering from chronic "surface-invasions." There would be opportuni of applying such inoculations in connexion with the treatment of bro chitis, ozaena, gleet, leucorrhoea, and those forms of the bacteruria[1] whi depend upon a bacterial invasion of the mucous membranes of the geni urinary tract. It would, of course, be necessary in each case, after det mining the particular species of invading micro-organism which was givi rise to trouble, to consider (1) the question of technique in connexion wi the preparation of the vaccines; (2) the question as to whether t particular invading micro-organism possessed any power of generalizi itself in the system; and (3) the question of the resisting and reacti power of the particular patient. In connexion with the estimation these last, the methods of estimating the bactericidal power of the blo which have been described by myself[2] and the method of estimati the phagocytic power of the blood which has been described by Maj Leishman[3] are, I think, capable of rendering services.

In bringing this communication to a close I desire to express r acknowledgments to Captain G. McIver C. Smith, I.M.S., for helpf collaboration in connexion with the study of Case 1. To my colleagu Major Leishman, R.A.M.C., I am indebted for manifold and unwearyi assistance and valuable criticism through the whole course of this inve tigation.

[1] Experiments in connexion with the application of inoculation in connexi with the last of these disorders are now in progress.

[2] *Proc. Roy. Soc.*, vol. lxxi, 1902.

[3] *Brit. Med. Journ.*, Jan. 11, 1902.

A Lecture on Therapeutic Inoculations of Bacterial Vaccines

And their Practical Exploitation in the Treatment of Disease.[1]

Delivered at the Medical Graduates' College and Polyclinic.

By A. E. Wright.

uence of Events after the Inoculation of a Bacterial Vaccine—Practical Importance of the Law of the Negative and Positive Phase in Connexion with Prophylactic Inoculations undertaken with Living or Sterilized Vaccines—Practical Importance of the Appreciation of the Law of the Negative and Positive Phase in connexion with the Therapeutic Inoculation of Sera derived from Animals vicariously Inoculated with Bacterial Vaccines—Therapeutic Inoculations of Bacterial Vaccines undertaken upon Patients already the Subjects of Bacterial Invasion—Treatment of Furunculosis, Sycosis, Acne, and localized Staphylococcus Infections generally by the Inoculation of a Staphylococcus Vaccine—Treatment of Cholelithiasis, Appendicitis, Colitis (also of Cystitis, Pyelitis and Endometritis, where these are produced by the Colon Bacillus) by the Therapeutic Inoculation of a Coli Vaccine—Cystitis and Localized Bacterial Infections of the Genito-Urinary Tract—Treatment of Tuberculosis by Therapeutic Inoculations of Tubercle Vaccine—Treatment of Bacterial Infections of Meninges of the Mucous Membranes of the Respiratory Tract, of the Middle Ear, of the Uterus, and of Joints by Therapeutic Inoculations of the Appropriate Vaccines—Summary.

E therapeutic method which I propose to consider with you to-day a method as yet almost unexploited. None the less it is a method ich is, if I am not mistaken, destined to revolutionize our ordinary actice in dealing with localized bacterial invasions. In dealing with ese our treatment has in the past consisted in making repeated applicans of antiseptics, or, in the case where this is impracticable, extirpating e seat of infection.

The time will come when, before embarking on either of these methods treatment, and above all before acquiescing in a policy of leaving the cterial invasion unchecked, an endeavour will be made in every case arrest the invasion and to prevent its recurrence by calling into action e forces of resistance which lie latent in the organism. *The physician the future will, I foresee, take upon himself the rôle of an immunisator.*

Before developing my ideas in a more concrete form, and pointing

[1] Reprinted from the *Brit. Med. Journ*, May 9, 1903.

out where the opportunities already lie for the exploitation of metho of immunisation, it will be essential for us to obtain a clear concepti of the immunising reaction which is initiated by the inoculation of vaccine. And we may group together under the appellation of vaccin attenuated living cultures of micro-organisms, sterilized cultures, ar derivatives of such cultures.

It will be convenient, at the outset, to discriminate from the mo complicated processes associated with actual disease the simpler reactio evoked by such vaccines. Let us note that in actual disease we ha to deal with a reaction of immunity hampered and often frustrated l processes of necrosis and cell degeneration induced by the action of tl bacterial toxins. In the case of properly-conducted vaccination proc dures, we have to deal with processes of immunisation uncomplicat and unfrustrated. It is with these immunising reactions—with thes if I may so denote them, physiological reactions—that we have he to concern ourselves. In dealing with them I shall not even attem to call up before you a mental picture of the hidden machinery in tl protoplasm which elaborates the products of immunisation which a found in the blood. You will already have been much wearied wi such attempts. I shall content myself with setting forth to you t sequence of events which supervenes when the machinery of the immun sation is set in motion by the inoculation of a vaccine.

Sequence of Events after the Inoculation of a Bacterial Vaccine.

The sequence of events after the inoculation of a bacterial pois was first clearly exhibited by Ehrlich[1] in connexion with a series

CURVE I.

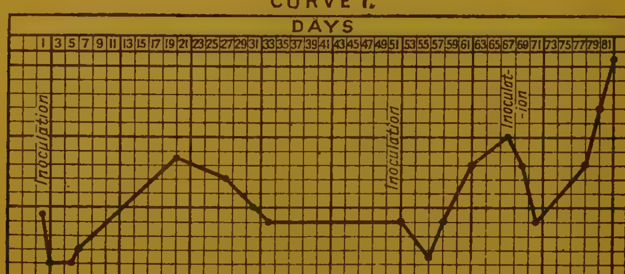


Fig. 1.—Curve of the immunisation reaction as obtained by measuring the conte in tetanus antitoxin in the milk subsequent to three inoculations of teta toxin.

inoculations of tetanus toxin undertaken upon a milch goat alrea previously immunised. The curve of immunity (Fig. 1) here reproduc from Ehrlich and Brieger's paper, discloses the content of the milk

[1] *Zeit. f. Hyg.*, vol. xiii.

tanus antitoxin at different intervals after the inoculation. The follow-g are the points to be noted :—

1. Immediately subsequent to the inoculation of tetanus toxin we ıve what we may conveniently speak of as a "negative phase" of ıe curve of immunity.

2. This negative phase is succeeded after an interval by a "positive ıase." In the case of the first inoculation here in question the climax the positive phase was marked by a duplication of the previous ıtitoxic power of the milk.

3. After a further interval the curve comes back to the higher base ıe which represents the quasi-permanent achievement of the immunisa-ɔn process.

The same succession of a negative and a positive phase is reproduced ter the second, and, so far as the curve was traced, after the third oculation.

The reaction of immunity was next studied by Salomonsen and adsen[1] in connexion with inoculations of diphtheria toxin undertaken ɔon a previously immunised milch mare. A reference to the curves Fig. 2 (page 230) which represent the anti-toxin content in the milk and ood respectively, will show that we have here again to deal with a gative and positive phase.

Similar curves have been obtained in connexion with other immunisa-ɔn processes, in particular by Bulloch[2] in connexion with the inocula-ɔn of ox-blood into rabbits, and by Morgenroth in connexion with the oculation of rennet.

I myself have, in a series of observations[3] made on men, obtained idence[4] of a negative and a positive phase of the bactericidal power the blood after typhoid inoculation. Two curves showing a negative d positive phase of bactericidal power after typhoid inoculation are esented in Fig. 3 (page 231). In later experiments undertaken by the me method I have obtained evidence of the maintenance of a higher se line two years after inoculation.

In a further research, dealing with the effects of anti-staphylococcus ɔculations[5] upon a series of men, evidence was obtained by me of e supervention of a negative and a positive phase of phagocytic wer[6] upon the inoculation of staphylococcus vaccines. I showed that association with the decline and rise of the phagocytic power, an gravation and improvement manifested itself in the clinical symptoms. typical curve is set forth in Fig. 4 (page 232).

[1] *Ann. de l'Inst. Pasteur*, 1887.
[2] *Trans. Path. Soc.*, 1902.
[3] *Lancet*, September 14, 1901.
[4] The method employed was that described by me in the *Proc. Roy. Soc.*, vol. i.
[5] *Lancet*, March 29, 1902 (pp. 199–226 *supra*).
[6] The method employed was that described by Leishman, *British Medical urnal*, January 11, 1902.

I think it may be taken as definitely established by the above tha we have to deal in connexion with every immunisation process with succession of a negative and positive phase. So far we have concerne ourselves only with a typical form of curve. We should, howeve lapse into fallacy if, carrying away in the mind's eye the features an general proportions of such typical curves as are shown in the figure above, we were to assume that these features and proportions must b reproduced in the case of every inoculation process. And similarl we should fall into error if we were to assume that a series of successiv inoculations would in every case, as in Fig. 1, lead to the achievement c a high base line of immunity.

CURVE 2.

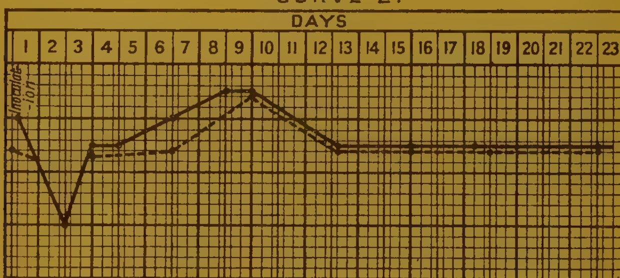
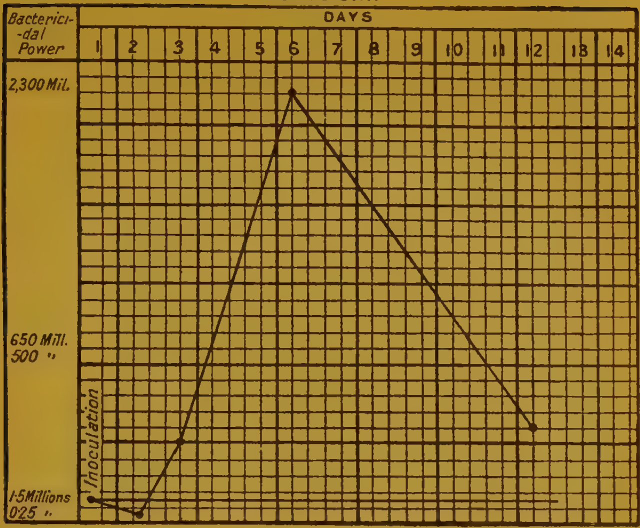


FIG. 2.—Curve of the immunisation reaction obtained by measuring the conter of the blood and milk of a mare in diphtheria anti-toxin after the inoculatio of diphtheria toxin. Unbroken line (——), antitoxin of milk ; dotted line (.......) anti-toxin of blood.

We shall best guard ourselves against errors such as those firs referred to if we consider the curves relating to the effect of anti-typhoi inoculation set forth in Curves 5 and 6 (page 233). In Curve 5 the negativ phase is suppressed, or speaking more strictly, the positive phase i found fully developed twenty-four hours after inoculation. In Curve the nadir of the negative phase is not reached till the ninth day afte the inoculations, the first evidence of a return in the direction of a positiv phase being obtained on the fifteenth day.

Let it be noted here that the differences in the duration of the negativ phase set forth in these traces are due either to comparatively sma differences in the dose of vaccine administered, or to idiosyncrasies o the part of the patients. If I may judge from experience of the result of the self-inoculation of a much larger dose of typhoid vaccine, and fror the results of blood examinations undertaken upon patients convalescen after very severe attacks of typhoid fever, it would seem to me that th incorporation or, as the case may be, generation in the system of exces sive quantities of typhoid poisons may be followed by a negativ phase of very many months' duration.

CURVE 3.A.



CURVE 3. B

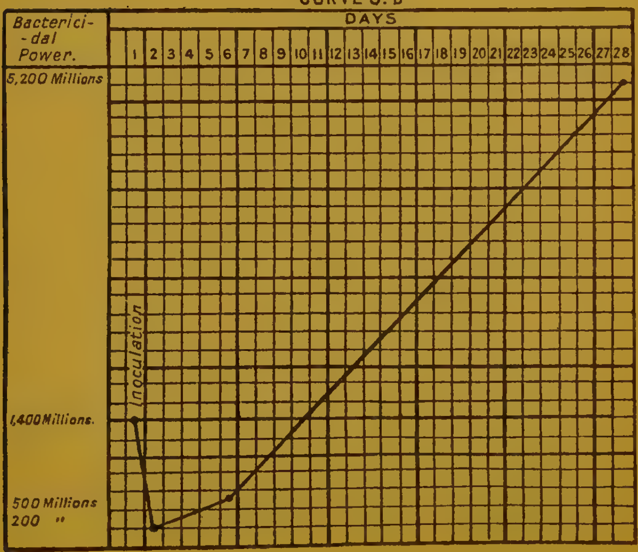


FIG. 3.—Curves (A and B) of the immunisation reaction obtained by measuring the bactericidal power of blood obtained from the finger before and after the inoculation of anti-typhoid vaccine. In Curve A all the measurements on the upper (positive) side of the base line have for convenience of exhibition been represented on a hundred-fold reduced scale.

CURVE 4.

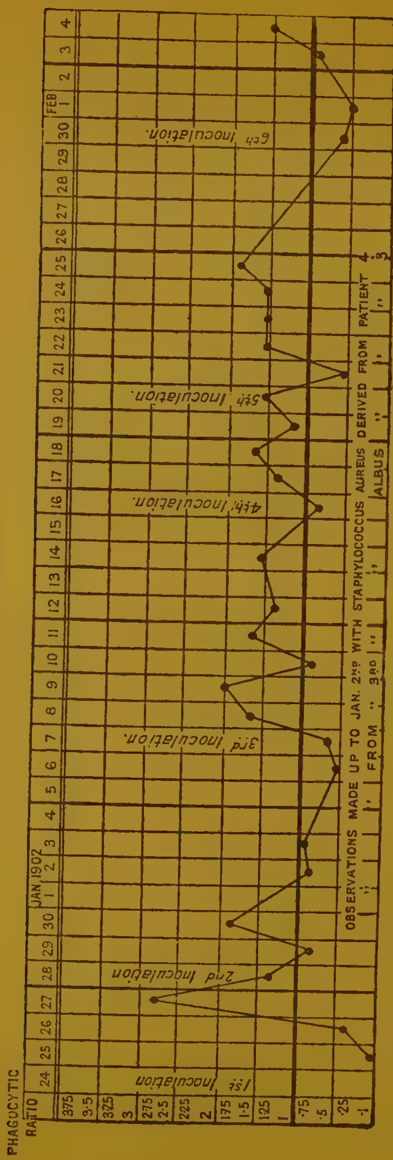


FIG. 4.—Curve of the immunisation reaction produced by a series of successive inoculations of a staphylococcus vaccine; the curve represents the phagocytic index of the patient's blood, the phagocytic power of the control blood being everywhere taken as 1.

What applies in the case of single inoculations applies with even greater force to the case where a series of successive inoculations is undertaken. It is of fundamental importance to recognize that the effect of every such series of inoculations is a cumulative one. It is

nulative in the sense of the negative phase and the poisoning of
 organism when the doses of vaccine are excessive, and when they
 upon the negative phases of the foregoing inoculations. And only
the case where the doses are properly adjusted and where each suc-
ding inoculation starts from a higher level attained by the previous
culation, will the effect of a series of inoculations be cumulative in the
ection of the positive phase[1] and in the direction of raising the organism
 a higher level of immunity.

CURVE 5.

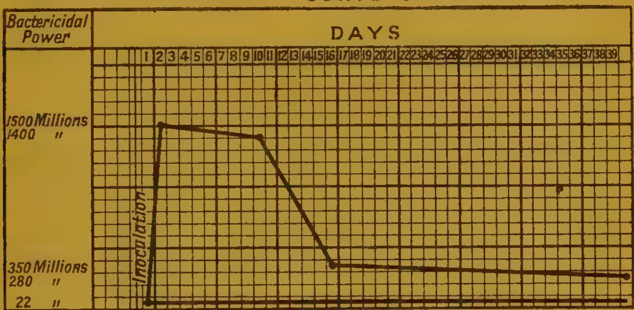


FIG. 5.—Curve of the immunisation reaction obtained by the inoculation of antityphoid vaccine, suppression of the negative phase.

You have now before you in outline what is known with regard to
 sequence of events following upon the introduction of a vaccine
o the organism, and with regard to what I may call the *law of the*
gative and positive phase and of the attainment of the higher base line. I
sire to insist upon the fundamental practical importance of this law
 connexion with every immunisation procedure. Let me, with a view
 illustrate its practical importance, deal, before I take up the subject
atter proper of my discourse, very briefly with the negative and positive
ase in connexion with the prophylactic inoculations of bacterial cultures,
d in connexion with serum-therapy.

CURVE 6.

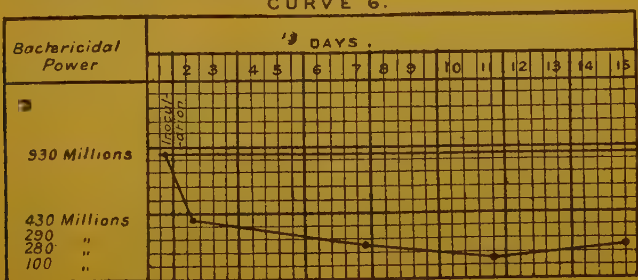


FIG. 6.—Curve of the immunisation reaction obtained by the inoculation of antityphoid vaccine, postponement of positive phase.

[1] *But vide p.* 273 *infra.*

Practical Importance of the Law of the Negative and Positive Phase i Connexion with Prophylactic Inoculations undertaken with Livin or Sterilized Vaccines.

In considering, in the light of individual results obtained in certai series of antityphoid inoculations undertaken in actually infected surround ings, the significance of the negative phase of bactericidal power afte antityphoid inoculation, I have called attention to the fact that th success of these prophylactic inoculations is imperilled where excessiv doses of vaccine are administered to patients in actually infected sur roundings, or immediately before transference to such surroundings. Basin myself upon information collected in India by the Indian Plague Com mission,[1] in connexion with Mr. Haffkine's antiplague vaccination, made a similar suggestion in connexion with this prophylactic inocula tion. And I have recently learned that the idea of a risk attachin to the inoculation of large doses of vaccine in infected surrounding suggested itself also to several observers in South Africa who had occasio to watch the effect of the antiplague inoculations there undertaken. I the suggestions made by me in connexion with antityphoid and anti plague inoculation are justified, we may not unreasonably expect t find indications of an increased susceptibility to small-pox in the perio supervening immediately upon the development of vaccinia pocks. I seems to me that such evidence can be found. In this connexion attentio may be drawn to the fact that a vaccine lymph which has altogethe failed to develop may, in the case where a more potent vaccine lymph is afterwards successfully inoculated, take on a typical development We can hardly explain this fact otherwise than by assuming that th resisting power of the patient which sufficed to hold in check the earlie applied and weaker vaccine is diminished or abolished under the influ ence of the toxins elaborated in the course of the development of th subsequently inoculated and more active vaccine.

Evidence of similar import, but from the nature of the case less con clusive evidence, is supplied by certain of the records of small-pox attacks and in particular by records of fatal small-pox attacks affecting thos who had been vaccinated immediately before. I would point out tha it is traditional in these cases to ascribe the small-pox attacks in questio to infection contracted previous to vaccination. It is also traditiona to vindicate this ascription by reference to the longer incubation o small-pox, overlooking the possibility that the incubation period may be shortened where the resistance of the organism is reduced.

A final word on this question will be appropriate. If the risks inci dental to the production of a negative phase attach, as I believe they do, to prophylactic inoculations in the case of all septicaemic diseases alike, it is obviously incumbent upon us neither to ignore nor to magnify these dangers, and, above all, to recognize that these risks can be mini-

[1] Report, Indian Plague Commission.

ed. Let it be observed that the risk of a negative phase comes ously into consideration only when excessive doses of vaccine are ployed, and when the prophylactic inoculations are undertaken in actual presence of infection. The remedy lies at hand. It lies in case where a sterilized bacterial culture is employed in the reduction he dose. It lies in the case of anti-small-pox vaccination in the reduc- of the number of insertions; in other words, in limiting the elabora- of the toxins by diminishing the area of skin surface employed for culture of the organism of vaccinia.

ctical Importance of the Appreciation of the Law of the Negative and Positive Phase in Connexion with the Therapeutic Inoculation of Sera derived from Animals vicariously inoculated with Bacterial Vaccines.

The principle of serum-therapy—that is, the idea of transferring to tients already the subjects of bacterial infection immunising sub- nces withdrawn from animals vicariously inoculated—appeals in very forcible way to the medical mind by the fact that it promises a ional treatment of all bacterial diseases, and by the fact that it has filled that promise in the case of diphtheria.

The prestige which it has derived from this signal success has led to e exploitation of the method of serum-therapy in connexion with nost every bacterial disease, and the time would appear to have arrived a general survey of the results. We can hardly fail to recognize that e new method of treatment has almost everywhere (I except, of course, e case of diphtheria) failed to do appreciable good. I would also ve you note that positive harm may upon occasion result from the sort to a serum. I may enforce this last statement by practical amples.

(1) There was submitted to the Indian Plague Commission a particular antiplague serum" intended for therapeutic administration. This rum, which was tested by me as a member of the Commission, pre- pitated the death of every plague-infected animal to which it was lministered, the acceleration of the fatal issue being in each case directly oportional to the dose of serum administered.[2] (2) In examining at etley a number of "antityphoid" sera with a view to selecting for nployment in South Africa any one that might seem calculated to be erapeutically useful, I found a particular "antityphoid serum" which ompletely abolished the bactericidal power which normal human serum erts upon the bacillus typhosus. (3) I have myself administered ith therapeutic intent to two Malta fever patients (personal friends) n "anti-Malta-fever serum," which produced unmistakeably toxic fects. The serum in question was a serum of my own preparation.) My friend Dr. Bulloch has had occasion to test on guinea-pigs the

[1] Report, Indian Plague Commission, chap. v.

effect of an "antistaphylococcus serum." The serum in question w found to exert a lethal effect in doses of 1 ccm. and over. (5) The sa has been observed by Marmorek in connexion with certain antistre tococcus sera, and instances are, I think, not infrequent—they ha certainly occurred within my experience—in which the administrati of "antistreptococcus serum" has seemed to produce toxic effec (6) A number of different scientific workers have, in endeavouring prepare an "antituberculous serum" by the inoculation of tubercul into horses, obtained a serum which produced on inoculation into ma and animals an unmistakable tuberculin reaction. (7) Lastly, Dunbar records the development of toxic effects in the case of a patient inoculat with an "anti-hay fever" serum.

These facts constitute, I think, striking confutation of the ide commonly entertained in our profession that all that is required for the pr duction of an effective therapeutic serum is the incorporation into a animal of progressively increasing doses of a bacterial culture, and th withdrawal of the serum ten days after the last injection. In oth words, the facts above enumerated enable us to realize that in defau of an active production of immunising substances on the part of th animal vicariously inoculated, the sera which are drawn off will inevitabl possess the toxic properties of vaccines originally inoculated. Ou awakening to this fact must be signalized by a re-examination of th whole question of serum therapeutics. That examination must b undertaken in the light of the facts which have been adduced above wit regard to the negative and positive phase.

Viewed from this point of view, the interpretation of the untowar results which followed the administration of the "antiplague serum" referred to above presents no difficulty. In point of fact, an inspectio of the horses from which the serum in question had been derived showe that the horses were still suffering from the fever and local disturbanc caused by the inoculation of the plague toxins. The serum had, i other words, been drawn off during the negative phase.[2]

Let it, however, be carefully noted that the passing off of the loca and constitutional symptoms does not in itself afford any guarante that the negative phase has given place to a positive phase. Persona experience in connexion with the inoculation of typhoid and Malta fever cultures into horses, the similar experience of others in connexion with the inoculation of other poisons into animals; and, lastly, experience of the effect exerted on human blood by antityphoid inoculation and by typhoid fever itself, make it certain that the persistence of the negative phase does not always betray itself by physical symptoms.

There is warranty for going further: there is, in fact, warranty for asserting that the persistence of the negative phase would in many cases be undetected by our present methods of testing sera.

[1] Dunbar, *Ursache u. specifische Heilung des Heufiebers*, 1903.
[2] Report, Indian Plague Commission, loc. cit.

Thus, for instance, the verification of the presence of agglutinins in serum which has been withdrawn does not in any way guarantee neutralization of the toxic substances which have been inoculated o the animal. Let it be observed in this connexion that the toxic ntityphoid" and "anti-Malta fever" sera referred to above possessed h—in the former, indeed, exceptionally high—agglutinating powers.

Again, experiments undertaken upon laboratory animals afford no urity for the absence of toxins from the sera under examination, ess the animals are equally sensible with man to the particular toxins ich are in question.

These points with regard to the difficulty of eliminating from use gative phase sera are, it will be seen, of altogether general application. they have been overlooked, this has no doubt been owing to the fact t it has been possible, in the case of the serum therapeutics of diphtheria, ing to the sensibility of the guinea-pig to diphtheria toxin, to eliminate toxic (negative phase) sera from use, and thus everything has in this pect here gone well.

The elimination of sera drawn off in the negative phase is, as considera-n will show, only one of the pre-conditions of a successful serum-therapy. further pre-condition of the successful exploitation of an antitoxic um is that there shall be transferred to the patient in the few cubic ntimetres of foreign blood which it is possible to administer a sufficiency antitoxin to neutralize at least a sensible fraction of all the toxin which being elaborated in the patient's system. In other words, it is essential at the animal which is selected for vicarious inoculation shall possess altogether phenomenal power of reaction with respect to the particular cterial poison employed. This condition, though it is satisfied in the se where horses are vicariously inoculated with diphtheria and tetanus xins, is not, so far as is at present known, satisfied in the case where imals are inoculated with any other poison.

Superadded to the difficulties already adverted to are other and greater fficulties which have reference to the therapeutic exploitation of anti-cterial sera. I refer to the difficulties created, on the one hand, by e circumstance that bactericidal elements disappear from the serum ery soon after the blood has been withdrawn, and on the other hand y the circumstance that these bactericidal elements may be incapable developing their effect when transferred from the vicariously inoculated nimal to the human patient.

Let me finally, before taking up the subject matter proper of my iscourse, try to sum up for you the situation.

The path of the immunisator who desires to proceed by the method of rum-therapy is bestrewn with all manner of formidable difficulties.

He must, in the first place, find methods which shall enable him to xclude from use sera drawn off during the negative phase.

He must, in the second place, achieve the hyper-immunisation of the nimal vicariously inoculated.

Lastly, in the case of anti-bacterial sera, he must find means of p serving the products of immunisation unaltered after withdrawal, and securing that they shall be operative within the system of his patient.

Therapeutic Inoculations of Bacterial Vaccines undertaken upon Patie already the Subjects of Bacterial Invasion.

If the situation as regards serum-therapy is as I have set it fo to you it manifestly behoves us to cast about and see whether there m not be a more excellent way. I venture to submit to your considerati —and here I arrive at last at the subject matter proper of this discourse the suggestion that that more excellent way may, perhaps, in many cas be found in the therapeutic inoculation of the patient with a bacter vaccine. Let me anticipate two *à priori* objections.

Let me make it plain, in the first place, that I am very far from su gesting the incorporation of additional bacterial toxins into a patie already the subject of a septicaemic disease or a serious bacterial into cation.

Such an inoculation could, I take it, serve no good purpo inasmuch as the bacterial elements similar to those that constitute t vaccine would in such a case already be circulating in the blood strea Furthermore, as you will immediately appreciate, the superadditi of additional bacterial toxins would inevitably prolong or reproduce t negative phase, if indeed it did not definitely turn the scale against t patient.[1]

The suggestion I make is that bacterial vaccines should be employ in the case where we have to deal with localized bacterial invasio associated with inflammation at the site of inoculation. The situati is here entirely different from that which has to be confronted in sept caemic diseases. On the one hand, the conditions are here already u favourable to the invasion of the blood stream by micro-organisms ; an there is, on the other hand, a considerable uncalled-on reserve of resistan on the part of the organism. We have to deal, in fact, with a situatio not altogether unlike that which obtains in the case of already partiall immunised animals, such as those employed by Ehrlich and Salomons in the experiments which we considered at the outset of this lectur Holding as we do in such a case a considerable balance in hand, we a in a position to confront without alarm the prospect of a possible temporar diminution of that balance. We are, in fact, in a position to adventur something for the sake of achieving afterwards a positive phase of increase resistance.

A further *à priori* objection which must be reckoned with is the follow ing : I may throw it into the form of a question. If it was legitimatel argued above that an inoculation of a bacterial vaccine would serv no good purpose in a case where the bacterial elements which call fort

[1] *But vide infra, pp.* 352–353.

immunising reaction are already circulating in the blood, does not s argument apply also to the case where bacterial toxins are orbed into the system from a localized seat of infection ? In point fact it seems not to apply. The explanation which suggests itself hat in such cases only the products of the metabolism of the bacteria, distinguished from the substances in the bacterial protoplasm which ke a production of antibacterial substances, are absorbed into the od stream.

What I picture to myself as the situation will perhaps appear to you a clearer light if I place before you the following series of facts :

The blood of the guinea-pig—and for our purposes we have to con- er only the male cavy—contains no spermatoxin ; in other words, exerts no poisonous effect upon the cavian spermatozoa. This is conformity with the circumstance that, while conceivably certain er metabolic products are absorbed into the blood from the testicle, spermatozoa themselves and their constituent elements are normally so absorbed. The conditions are here presumably analogous to se which obtain in the case where bacteria are cultivating themselves ally in the organism.

If we now, after extirpating one of the guinea-pig's testicles, inoculate n subcutaneously with a suspension of spermatozoa obtained by king an extract of the extirpated testicle, we find that his blood develops rmatoxic properties, and that when brought into contact with actively ving spermatozoa it immediately arrests their movements. Without ssing home the analogy, we might on a similar principle expect to luce an elaboration of antibacterial substances by inoculating a bacterial ture into a patient already the subject of a localized infection.

Having, so far as I can, disposed of the *à priori* objections which ght suggest themselves in connexion with the proposed method of rapeutic inoculation of bacterial vaccines, and having given you, I pe, a certain insight into the principles of the proposed method, I will w ask you to consider where the opportunities lie for the application the method. I may appropriately begin by discussing those thera- utic inoculations of which I have already had some experience.

eatment of Furunculosis, Sycosis, Acne, and Localized Staphylococcus nfections generally by the Inoculation of a Staphylococcus Vaccine.

There can, I think, be no doubt that in acne vulgaris, furunculosis, d most cases of sycosis we have nearly always to deal with an invasion the skin follicles, or, as the case may be, of the subcutaneous tissue the staphylococcus pyogenes. In almost every case the suppurating eas yield the staphylococcus in pure culture. This, however, is only e of the factors in the causation of these disorders.

Another factor is revealed by an investigation of the phagocytic wer of the blood. This examination has, in all the cases I have

as yet dealt with, revealed a defective power of phagocytosis w respect to the staphylococcus. We have, it seems to me in the abo taken in connexion with the extreme chronicity, tendency to relap and comparative non-infectivity which characterizes these disorde clear indications of the line of treatment that ought to be adopted. ought to aim primarily at the immunisation of the patient against t staphylococcus.

I have elsewhere[1] given an account of six consecutive cases wh were treated by the inoculation of a staphylococcus vaccine, and ha set forth the details of the blood examinations, and in association w them the clinical result. In two of these cases marked, but only te porary, improvement was achieved; in the four others a comple and very rapid cure was effected. The cases in which the improveme was only partial and temporary were—a case of boils and irrita pimples occurring in an elderly lady as a sequel of a severe operatio and a case of extensive sycosis and eczema barbae dating back eig years to almost the period of puberty. In both of these cases the appears to have been a deficient power of response to the inoculatio

Among the cases cured, the most conclusive from the point of the pr vious chronicity of the morbid process, the severity of the sympto and the subsequent following up of the case, was that of an officer wh subsequent to a septic infection which eventuated in peritonitis, h for a period of seven years been a victim to sycosis, eczema barbae a tarsi, styes in the eye, and deep and superficial furuncles. All the troubles disappeared after three successive inoculations of staphylococc vaccine. His face had remained absolutely free from eruption wh the patient, eighteen months after the date of his original first inoculatio presented himself for reinoculation on account of the development two incipient boils, these being the first which had developed since unde going the treatment. The reinoculation was effectual in checking a further relapse.

It may be added that, owing to the relatively small toxicity staphylococcus cultures, the therapeutic treatment by staphylococc vaccine involves comparatively little discomfort. The discomfort i at any rate, absolutely trivial in comparison with the mental sufferin associated with disfigurement due to acne or sycosis, and the physica evils associated with the reiterated application of antiseptics, epilatio and the fomentation and lancing of furuncles.

If staphylococcus vaccine should continue to approve itself usef in the treatment of localized cutaneous invasions of the skin, this woul manifestly pave the way for further therapeutical applications in co nexion with staphylococcus invasions of wounds, and open ulcers an granulating surfaces generally.

[1] *Lancet*, March 26, 1902 (pp. 203–223 *supra*).

atment of Cholelithiasis, Appendicitis, Colitis (also of Cystitis, Pyelitis, and Endometritis where these are produced by the Colon Bacillus) by the Therapeutic Inoculation of a Coli Vaccine.

The colon bacillus, even if we consider it quite apart from its near geners, the dysentery bacillus, the Gaertner bacillus, and the typhoid illus, all of which, like the colon bacillus, affect by predilection the er portion of the ileum and upper end of the large intestine, is respon-e for an altogether astonishing amount of human ills. The rôle ch the colon bacillus plays in connexion with internal surfaces is, fact, not less predominant than that played by the staphylococcus connexion with external surfaces and wounds. And the gravity the local inflammatory processes which result from the inroads of colon bacillus is infinitely greater. These derive their gravity, first, n the importance of the organs which are involved; secondly, from anatomical relations of these organs with the peritoneum; thirdly, n the practical impossibility of effectively applying antiseptics; , fourthly, from the very serious surgical procedures which are required he case where the localized invasion culminates in the production ous and shows a tendency to generalize itself. All these are considera-is which plead in favour of dealing with colon infections by the method mmunisation.

Time will not permit of my referring to more than one or two of the ny problems which open out before us when we take into considera- the possibility of dealing with colon bacillus infections or by the hod just suggested. And you will understand that even if time did mit, the limitations of my knowledge in this imperfectly explored field ld very soon impose a restraint upon me. It may, however, not altogether unprofitable to suggest to you in connexion with this stion certain lines of thought.

Cholelithiasis.—Evidence amounting now almost to demonstrative of has been accumulating showing that the formation of gall stones ependent upon bacterial invasion of the gall bladder, just in the same as the formation of the ordinary phosphatic calculus in the bladder ependent upon a bacterial invasion. It has further been established the colon bacillus is the particular micro-organism which is respon-e for the formation of gall stones. Not only is this micro-organism nd in pure culture in the gall bladder in most cases of cholelithiasis, it is found also in the interior of practically all recent biliary concre-s. Furthermore, it has been shown that gall stones can be experimen-y produced by a direct inoculation of the colon bacillus into the gall lder. And, lastly, it is interesting and important to note that Professor ghan Harley has recently found that ordinary gall stones are spon-eously dissolved when they are introduced into the gall bladders of s, while no such solution occurs when they are introduced in associa- with pus derived from the inflamed gall bladder of man.

It would seem, in view of the above, that the treatment of choleli asis ought to be directed to the immunisation of the patient agai the colon bacillus. We can see that it is within the bounds of possibi that the colon bacillus infection might in this way be checked, and t the gall stones might be dissolved. Even if these ends were not achie the immunisation would at least prepare the way for surgical p cedures by diminishing the attendant risks. Let us note with regard the efficacy of these surgical procedures that the removal of the g stones would not by itself make an end to the evil.

Appendicitis.—More important by far than the pathology of g stones is the pathology of appendicitis. Without for a moment c tending that the colon bacillus is the only micro-organism involv in the etiology of appendicitis, it appears to me to be beyond doubt essentially important agent. I find, for instance, on looking up records of bacteriological examinations undertaken in the surgi theatre or, as the case may be, the *post mortem* room at St. Mary that the colon bacillus was in eight successive cases obtained in p cultivation from the pus, and that it was obtained in pure culture fr the contents of the caecum or appendix in five out of the six cases which cultures were made from the contents of the intestinal canal.

Here again it would seem to me that treatment ought to be direct to the immunisation of the patient against the colon bacillus. Immuni tion procedures might, it seems to me, appropriately be undertaken the one hand with the design of preventing a recurrence and avoidi operation, and on the other hand with the design of preparing the patie for operation in those cases where surgical procedures are postpon until after the subsidence of the symptoms of inflammation. The ris of inducing peritonitis which are incident to the necessary breaki down of adhesions and opening up of old foci of infection might conceivab be appreciably diminished by these means. The results which have be obtained in man by anticholera and antityphoid inoculations, and anima by anticoli inoculations, would seem to show the possibility of effecti immunisation against bacterial infections proceeding from the intestin canal.

Cystitis and Localized Bacterial Infections of the Genito-urinary Tra

It is hardly necessary to do more than advert to the frequent associ tion of cystitis and pyelitis with an invasion of the urinary tract by t colon bacillus. It is perhaps less well known that certain cases of end metritis are associated with an invasion of the uterus by the same micr organism. It would seem possible that patients, the subjects of the infections, might be advantageously treated by inoculations of c vaccine. In two cases in which I have already carried out such inocul tions, the patients have claimed that they derived benefit from them. was, however, unable to detect any objective evidence of improveme

bacteriological examination of the urine. It is obvious, however, t the number of coli bacilli in the urine is not necessarily a correct ex of the condition of the local inflammation process in the wall he urinary tract.

atment by Tuberculosis of Therapeutic Inoculations of Tubercle Vaccine.

A few words by way of introduction to the discussion of this question perhaps serve to place matters clearly before you. The treatment uberculosis by tuberculin as originally introduced by Koch was in ntion a method of *toxi-therapeutics.* The tuberculous toxins which e inoculated were intended to compass the expulsion and destruction he invading tubercle bacilli by effecting necrotic and degenerative nges in the foci of infection. It is now universally recognized that therapeutic principle was fundamentally erroneous. It is unnecessary ay more, except that the results were untoward and that the necrotic, enerative, and inflammatory changes which were effected in the foci of ction contributed, in some cases, to the dissemination of the tubercle lli in the infected system.

Of recent years modifications have been made by Koch in the prepara- of the tuberculin, which have as their result the elimination of the e soluble toxins and the incorporation into the menstruum of the soluble elements of the bacterial protoplasm. In short, modifications e been made which bring tuberculin (the so-called new tuberculin or . and T. R. tuberculins) more into line with our ordinary bacterial ines.

Following upon these changes a complete change of policy has been ly inaugurated. The tuberculin inoculations have been definitely di- d of the character of toxi-therapeutic inoculations, and have been itely invested with the character of therapeutic inoculations of a tubercle ine designed to call forth an antibacterial reaction in the organism.

As soon as we consider the antitubercle inoculations from this stand- t, everything that has occurred in connexion with them becomes ediately intelligible. We appreciate, in the first place, that the ward results which so often supervened upon the inoculation of Koch's nal tuberculin were, in so far as they were not the result of the nerative and inflammatory changes above referred to, imputable he inappropriate character of the vaccine, and often, no doubt, to induction of a cumulative negative phase under the influence of essive inoculations uncontrolled by intermediate blood examinations. appreciate further that the outlook for a useful exploitation of thera- ic inoculations for tubercle is more hopeful now that we have at osal a vaccine which contains in it elements derived from the erial protoplasm ; and now that we have at disposal in the tuberculous n-sedimentation reaction a method by which we can obtain some

information of the progress of the reaction of immunisation in the patie
system. I would submit to you that since we now need no longer w
entirely in the dark, we ought cautiously to exploit in the treatm
of localized tuberculous affections the tubercle vaccine which we ow
the ever fertile labours of Koch.

Treatment of Bacterial Infections of the Meninges, of the Mucous Me branes of the Respiratory Tract, of the Middle Ear, of the Ute and of Joints by Therapeutic Inoculations of the Appropriate Vacci

Before proceeding to summarize the general results we have arri
at, I may perhaps take an opportunity of explaining that the therapeu
method which is here in question is capable of an even more exten
application than that sketched out above. I foresee that if the princ
of the therapeutic inoculation of bacterial vaccines establishes itself,
I do not doubt that it will in the future, as an approved method for
treatment of chronic and recurrent inflammation processes, it will
exploited also in connexion with chronic and recurrent meningeal dise
(I have in view here in particular certain cases of infantile paralys
middle-ear disease, chronic bronchial and uterine catarrh, and chronic jo
affections. Every such case would, of course, be submitted to bact
ological examination with a view to the determination of the determin
cause of the infection, and the employment of the appropriate bacte
vaccine. In considering such a treatment in connexion with a mening
infection there may be borne in mind the fact that we have, in
case of the anti-rabies inoculations, a demonstration of the possibil
of checking, by the means of subcutaneous therapeutic inoculation o
vaccine, the spread of infective micro-organisms in the cerebro-spi
system. Similarly, in considering a possible application of bacte
vaccine in connexion with inflammation processes affecting the muco
membranes of the respiratory system, we may derive encourageme
from the results obtained by Professor Dunbar in connexion with inocu
tion against hay fever. For an elaboration of an antitoxin against t
pollen toxin in the organism of animals has been achieved by him, a
he has obtained indications of a similar production of antitoxin in t
organism of a susceptible human patient.

Summary.

Let me, finally, recapitulate to you the conclusions we have arriv
at in the course of our study.

1. We have seen that we have in connexion with every immunisati
process a sequence of *negative* and *positive phase* followed in the case whe
the inoculation is successful by the maintenance of a *higher base li*
of immunity.

2. We have seen that the inoculation of an excessive dose may invol
a risk, in particular the risk of an undue prolongation of the *negat*
phase.

3. We have seen that the inoculation of a series of doses of a vaccine l, in the case where the inoculations are uncontrolled by intermediate od examinations, involve the possibility of the production of a *cumula-: negative phase.*

4. We have seen that the *cumulative positive phase* which is a desider-m either in itself or as leading to the maintenance of a high base e of resistance, is achieved only when the successive doses are properly usted and when the inoculations are appropriately interspaced.

5. We have seen that the success of a prophylactic inoculation process y be imperilled where sequence of negative and positive phase, and cumulative effect of successive inoculations, is not taken into con-eration.

6. We have seen that the success of serum-therapy in diphtheria and comparative failure in the case of other diseases is explained by the t that in the first case we are able to secure the elimination of all nega-e phase blood, and we are able to induce in the vicariously inoculated mals a cumulative positive phase of absolutely phenomenal dimensions. the case of other diseases we have been unable to secure these pre-uisites of a successful serum-therapy.

7. We have seen that in the case of patients who, though suffering m localized bacterial invasions, are possessed of a considerable balance resisting power, it is possible without risk to undertake therapeutic culations of bacterial vaccines, provided always that the results of se inoculations are controlled by subsequent blood examinations.

Let us realize, in conclusion, that when all has been done that can done in the way of guarding a patient against the risks attaching the negative phase, the success of a therapeutic inoculation cannot be ranteed. The success must in each case depend upon the power of ponse which is possessed by the individual. In the case of a particular ient that power of response may fail us. It may fail us also in con-ion with particular bacterial infections. But it may be predicted t success will in some cases be achieved. It seems to me, for ance, that it will certainly be achieved in the case of simple staphy-occus infections occurring in the young and otherwise robust. It ms to me very probable that it will be achieved also in the case of tain coli infections.

On the Treatment of Acne, Furunculosis, an Sycosis by Therapeutic Inoculations c Staphylococcus Vaccine.[1]

By A. E. Wright.

C'a été une chose curieuse que la faillite des antiseptiques dans le traitement des malad dermatologiques parasitaires. On fondait sur eux des espérances colossales, n'ont presque rien donné.—Sabouraud.—*Bulletin de l'Institut Pasteur,* 190 p. 286.

Three and a half years ago I was confronted with the following situ tion: I was consulted by a patient, forty years of age, who was a pre to chronic staphylococcus invasions, and had for seven years suffere from severe and constantly-recurring furunculosis, complicated b sycosis, eczema barbae, styes in the eyes, and eczema tarsi. The boil which occurred all over the body, occurred in two varieties—sma superficial boils seated generally in the skin of the neck and face, an larger deep-seated boils occurring in the subcutaneous tissue of ever region of the body, more particularly about the nates and thighs. Hi history was as follows.

Seven years previously, when engaged in clearing out a tracheotom tube which had been removed from a patient who had been operated upo for acute laryngitis, he accidentally inoculated himself in the forefinge with some of the septic material, and in spite of three deep incision made successively into the finger and palm of the hand, the infectio spread upwards to the axilla, giving origin there to a bubo, and thenc onwards into the blood stream, setting up high fever and septic peritonitis The susceptibility to staphylococcus invasion had manifested itsel immediately after the septic attack.

Immediately before the date at which the patient consulted me had been engaged in following out the changes which are produced in the blood after the inoculation of antityphoid vaccine.[2] I had ascer tained in the course of that inquiry that the inoculation of a bacteria vaccine is followed, first, by a negative phase of diminished bactericida

[1] Reprinted from the *Brit. Med. Jour.*, May 7, 1904.

[2] On the Changes effected by Antityphoid Inoculation in the Bactericidal Power of the Blood, with Remarks on the Significance of these Changes, *Lancet*, September 14, 1901.

ver, corresponding, no doubt, to the period of application of the vaccinal nulus ; secondly, by a positive phase of greatly increased bactericidal ver, that is, a period of active response ; and lastly, after the remission he stimulus, by a comparatively durable period of increased resistance. ad further determined that the negative phase effect is directly depen-t with respect to duration and intensity upon the dose of the vaccine, n negative phase being of only short duration when the dose of the cine inoculated is small, and where the constitutional symptoms duced are slight. Reflecting upon these facts, it occurred to me to ak through conventional restrictions, and to exploit bacterial inocu-ons, not alone for the purpose of prophylaxis but also therapeu-lly, in the case of patients suffering from localized bacterial invasions. ing effect to these considerations, I proceeded in a tentative manner esting in each case the effect produced upon the blood by each arate inoculation—to experiment on the possibilities of applying culation of a staphylococcus vaccine in the treatment of furunculosis, osis, and acne. I published my first results obtained on a series of cases in the *Lancet* for March 29, 1902. I set forth in the paper in stion in detail the method employed for the preparation of the vaccine, l the course of the reaction of immunity as indicated by the phagocytic ction of the blood before and after inoculation. It emerged very arly that we have, in the case of the inoculation of staphylococcus ccine, to deal with a precisely similar sequence of negative and a itive phase as in the case of antityphoid inoculation ; and further, t, as in the case of antityphoid inoculation, the duration of the gative phase varies with the quantum of the vaccine inoculated ; tly, that the attainment of the desired clinical result depends on the nulation of the effects of a series of properly interspaced inoculations. e clinical results achieved in the six cases here in question were very l of promise. The cases that were treated were, first, the case of vere and multiform staphylococcus invasion just described ; secondly, case of very severe sycosis, which had lasted for a period of years, d which had been intractable to all treatment ; thirdly, a case of runculosis in an old lady which had followed upon a surgical opera-n : fourthly, a case of acute furunculosis in a medical student ; fifthly, ase of mild sycosis ; and, lastly, a case of irritable furuncular pimples the back of the neck.

I have since the date of the publication of the paper in question eated by the method of inoculation a further series of cases of aphylococcus invasion. These cases include : One case of severe neralized staphylococcus invasion characterized by boils, paronynchia, d severe acne ; three cases of severe and long-continued sycosis barbae, nich had proved intractable to treatment by antiseptics ; a further se of sycosis barbae, which had been treated by epilation for many ars, six cases of moderate furunculosis ; and seven cases of severe d very chronic acne. In nearly all these cases I have plotted out

in the form of a curve the changes produced by inoculation in t phagocytic power of the blood.

The following are brief particulars of some of the cases.

Case 7.—The patient is a medical man, the curator of a path logical museum. He suffered more or less continuously for years fro boils in all parts of the body, from severe facial acne, and from paronych We have here evidence of a susceptibility to every form of staphylococc invasion. In April, 1903, after removal of four finger-nails for pa onychia, he applied to me with a view to subjecting himself to staph lococcus inoculations. At the time the patient was suffering also fro "a large crop of boils upon his neck." He writes, "after the fir inoculation, these quickly aborted, and I remained in very good heal until August, when a large furuncle formed in my right buttock." Th furuncle aborted after undertaking a second inoculation. The patie reports (January, 1904) "that the facial acne has been considerab better since the inoculation, but I am," adds he, "by no means cure as I had another furuncle in the loin a few days ago, but I must sa that I have had great benefit from the inoculations." Owing to th fact that the patient was treated in the provinces, no data are he available with regard to the condition of the blood before and aft inoculation.

Case 8.—Patient, a labourer, forty years of age, the subject c sycosis, gives the following history: He suffered as a child from sever pustular eczema and from deep-seated suppuration behind the ea which continued for years and has left very deep scars. He has als suffered from time to time from boils and from a discharge from th ears. We have here probable evidence of a lifelong susceptibility t staphylococcus invasions.

Five months ago his head became very scurfy, and the inflammatory affection passed down from the head to the parotid region, and finally to the beard and to the whole hairy surface of the face and chin. The patient now presented himself for treatment as an out-patient in the Skin Department of St. Mary's Hospital, and was treated by the X rays and by antiseptics for two months without any improvement. It was now determined to try the effect of staphylococcus inoculations.

Condition of the Patient when the First Inoculation was undertaken. —November 2, 1903. The patient was suffering from very aggravated pustular sycosis, each hair of the beard being surrounded by pus, while there was considerable induration and furuncular inflammation below the angle of the jaw and over the whole anterior surface of the neck. The patient's phagocytic index, taking unity as representing the phagocytic index of the normal blood, was 0·48. The patient was inoculated with a quantum of sterilized staphylococcus culture which contained 2,500 million staphylococci.

November 9. Sycosis is astonishingly improved. The general health he patient is also much better, and the pain has entirely disappeared. e phagocytic estimation miscarried. The patient was inoculated with uantum of sterilized staphylococcus culture containing 5,000 millions staphylococci. Cultures obtained from the remaining pustules lded pure cultivations of the staphylococcus aureus.

November 16. Improvement has been continuous, there being now y few pustules. Acute weeping eczema, has, however, supervened r the whole region of the head. The patient was treated for this dition by Dr. Graham Little by an application of carbolic oil. Phago- ic index, 1·21.

November 19. Improvement still continuous, so far as the pustulation the beard is concerned. The patient still suffers from weeping ema over the whole region of the scalp. Lotio plumbi was substituted the carbolic oil. Phagocytic index, 1·13. The patient was rein- lated with a quantum of sterilized culture which was derived from own staphylococcus,—the quantum inoculated corresponding to 00 millions staphylococci.

November 26. Pustules on the face, head and arms, had now com- tely disappeared, and the eczematous condition is nearly well, but a d deal of redness persists about the skin and cheeks, and the epidermis everywhere scaling off. Phagocytic index, 2·1.

November 30. A few trifling pustules are still to be seen on the e, but much less general congestion. Phagocytic index, 1·92.

December 8. The patient is now almost quite well, with the exception a few superficial pustules. Phagocytic index, 2·7. The patient s inoculated with a quantum of vaccine made from his own staphylo- cus containing 7,500 millions of staphylococci.

December 14. The patient is practically well. Phagocytic index, 0·92.

December 17. The patient is about to resume work. Phagocytic ex, 1·85.

Subsequent History.—The patient, who was all but completely cured December, relapsed as a consequence of alcoholic excesses at Christ- s. He is now, after a further series of inoculations, again well.

Case 9.—The following case was inoculated and examined by my nd, Captain Stewart R. Douglas, I.M.S., in the Royal Victoria spital, Netley. The patient, Private C., was affected with sycosis the beard. Two years ago, when on active service in South Africa, a p of small pimples developed on his chin and spread over the whole face of his face and jaw. The patient has been in hospital under atment for seventeen months, but his condition has undergone only nporary improvement.

May 15, 1903. Phagocytic index, 0·8.

May 23. Phagocytic index, 0·7. Inoculated with a sterilized

staphylococcus culture derived from his beard. The quantum inoculated corresponded to 2,500 millions staphylococci.

May 25. Phagocytic index, 1·1.

May 28. Phagocytic index, 1·2.

May 28. Phagocytic index, 1·2. Reinoculated with a quantum of the same vaccine containing 5,000 millions of staphylococci.

May 29. Phagocytic index, 0·64.

June 2. Phagocytic index, 2·6.

June 3. Phagocytic index, 1·9.

June 4. Phagocytic index, 1·45.

June 6. Phagocytic index, 1·2.

June 7. Phagocytic index, 1·5.

June 9. Phagocytic index, 1·2.

After the first inoculation, and contemporaneously with the rise in the phagocytic curve, the number of the pustules was considerably diminished. After the second inoculation the patient was practically well. Captain Douglas's further observations were broken off by his departure from Netley.

Case 10.—The patient, a medical student from a Northern university, had been for years the victim of severe sycosis complicated by abscesses in the parotid region. He had been treated in a very systematic manner by epilation, and there was no longer any active sycosis in progress. There remained, however, considerable congestion and induration in portions of the previously affected area.

Phagocytic index, 0·73.

The patient was inoculated with a quantum of staphylococcus vaccine containing 2,500 millions staphylococci.

June 3, 1903. The symptoms after inoculation have been very slight. Phagocytic index, 2·25.

June 4. Phagocytic index, 1·2.

June 5. The patient was reinoculated with a quantum of vaccine corresponding to 5,000 millions staphylococci.

June 6. Patient has considerable reaction and local pain.

Phagocytic index, 0·57.

June 12. Phagocytic index, 0·79.

June 15. Phagocytic index, 1·2.

June 17. Phagocytic index, 1·1.

Reinoculated with 2,500 millions staphylococci.

June 20. Phagocytic index, 0·8.

June 30. Patient expresses himself as convinced that his condition is much improved.

Phagocytic index, 1·06.

July 3. Reinoculated with 5,000 millions of staphylococci derived from the culture which was obtained before the first inoculation.

July 6. Patient is still suffering from considerable constitutional disturbance.

Phagocytic index, 0·34.

July 8. The constitutional disturbance has passed off. Phagocytic ex, 1·75. Treatment suspended, the congestion and induration ring practically disappeared.

January 4, 1904. Patient presented himself, with a history of some urrence of his facial trouble, from which he had been free for five nths. After two further inoculations with a vaccine prepared from staphylococcus isolated from his beard six months ago the inflamma-y trouble again completely subsided.

Case 11.—June 12, 1903. The patient, a medical man, presented nself for treatment with the following history. He suffered from the t attack of boils in June, 1897, after exhausting outdoor work in nexion with midwifery. His attack lasted for about six weeks, and eries of about a dozen boils developed. During the next eighteen nths he suffered from several attacks of boils, but the details of these acks had faded from his memory. At the end of 1900 and spring 1901 he suffered from a number of recurrent attacks of furunculosis, eral of these lasting well over a month.

In 1902, in February, he suffered from a very aggravated boil, and 1903 from two or three bad attacks. Between June 2 and 12, the on which the patient presented himself for treatment, nine boils appeared in succession on his neck.

June 12. Phagocytic index, 0·87.

The patient was inoculated with a quantum of sterilized culture con-ning 500 millions staphylococci.

June 13. Phagocytic index, 0·73.

June 15. Phagocytic index, 0·9.

June 18. Patient after wearing a collar for the first time on June developed two further boils. One of these aborted.

June 20. Phagocytic index, 1·02.

The patient was reinoculated with 750 millions staphylococci.

June 24. Succeeded in wearing a collar for a few hours in the evening.

June 25. Developed a small boil, which cleared up on the 28th.

June 27. Phagocytic index, 1·2.

June 28. A spot on the right eyebrow, which did not suppurate.

July 3. Phagocytic index, 1·1.

Patient was reinoculated with a vaccine made from a staphylococcus ained from his boil. The constitutional reaction which resulted s somewhat more severe than on previous occasions.

July 8. Phagocytic index, 1·3.

Subsequent History.—Patient remained perfectly free from boils for er two months.

In October, after three weeks of sleeplessness, patient developed a all boil behind angle of jaw and stye in the eye.

In January, 1904, a small pustule appeared on the neck; with these

exceptions patient has had no recurrence of his trouble. He expre himself as convinced that his period of insomnia would, except for inoculation, have been followed by an aggravated attack of boils.

CASE 12.—The patient, a medical man, aged fifty-five, applied treatment, having suffered from boils on the back of the necl frequent intervals for three or four years. He has always been m or less subject to this trouble.

December 4. Phagocytic index, 0·79. Patient was inoculated v a quantum of staphylococcus vaccine corresponding to 2,500 milli staphylococci.

December 10. Patient was reinoculated with 1 ccm. staphylococ vaccine.

December 12. A boil has begun to develop in the neck.

December 17. The boil under the ear, which would in ordin circumstances have suppurated and sloughed, is now aborting. Pha cytic index, 1·56.

Subsequent History.—Patient has had no recurrence of boils.

CASE 13.—July 14, 1903. Patient, a healthy man, aged twenty-fo who was in training for a boat race, developed a boil on his gluteal reg three weeks ago, and is now suffering from a crop of boils on the ne Phagocytic index, 0·84. Patient was inoculated with a quantum vaccine corresponding to 2,500 millions staphylococci.

July 17. Boils have improved. Phagocytic index, 0·88. Patie was reinoculated with 2,000 millions staphylococci.

July 20. Boils are nearly well. Phagocytic index, 1·3.

July 25. Phagocytic index, 1·9.

July 31. Phagocytic index, 1·95. Patient soon afterwards relaps and did not come up for further treatment.

CASE 14.—Patient, a professional man, aged thirty-two, present himself for treatment on November 25, 1903, having suffered almo without interruption since last May with painful boils on the nate which in each case suppurated. The patient gives a history of sycos ten years ago.

November 25. Phagocytic index, 0·87. Inoculated with 2,50 millions staphylococci.

December 4. Patient has developed another small boil in the glute region. Phagocytic index, 0·91. Patient reinoculated with 5,00 millions staphylococci.

December 17. The boil referred to above has aborted withou suppuration. Phagocytic index, 0·94.

Subsequent History.—Patient remained free from his trouble for thre and a half months. He then developed a boil in the neck, and wa reinoculated with 2,500 millions staphylococci.

Case 15.—Patient, a professional man who has been invalided me from the East for general debility and boils, presented himself : treatment with a boil in each axilla.

October 8, 1903. Phagocytic index, 0·49. The patient was inocu-ed with 2,500 millions staphylococci.

October 15. Patient is much improved in health. The boils in the illa have aborted without suppuration. He was reinoculated with 00 millions staphylococci.

November 1. Phagocytic index, 0·95.

Subsequent History.—Patient is in good health, and has had no rther boils.

Case 16.—Patient, a medical student, aged twenty-four, with a long story of acne and boils. Boils have occurred in every region of the dy, they are at present quiescent. The face is a mass of scars from d acne. There is at present in comparison with what there has en, a trifling amount of active mischief.

December 1, 1903. Phagocytic index, 0·82. Inoculated with a antum of vaccine corresponding to 1,000 millions staphylococci.

December 8. Patient reports himself much better. Phagocytic dex, 1·7. Reinoculated with 5,000 millions staphylococci.

December 18. Patient's appearance has improved in a wonderful anner, every trace of pustulation having disappeared.

April 25. Patient has, as a result of working for an examination, gun to relapse. He was reinoculated with 2,500 millions staphylococci.

Case 18.—Patient, twenty-five years of age, is engaged in the City. as been the victim of pimply acne for many years. At present there e very few indications of suppuration. Phagocytic index, 0·54. oculated with 2,500 millions staphylococci.

December 2, 1903. Patient's condition seems to be a little ameli-ated, patient himself being uncertain whether this is to be attributed the inoculation or to an ordinary remission of his trouble. Phagocytic dex, 1·0. Reinoculated with 5,000 millions staphylococci.

December 11. No great change. Face appears somewhat clearer. agocytic index, 2·1.

Subsequent History.—Patient presented himself two months after ith an almost perfectly clear complexion. He expressed himself as tisfied of the efficacy of the inoculations.

Cases 17, 18, 19, and 20.—These were all cases of aggravated and very ronic acne, affecting respectively a young medical man, a young labourer, spinster lady of forty-five, and a married lady of thirty-five. The st two cases were characterized by marked pustulation and extensive arring over the face, the upper part of the chest, and the upper half the back. In the third case the chief feature of the condition was e reddening and development of indolent pimples over the region of

the nose and chin. The fourth case was characterized by the deve ment of marked induration round the papules, in particular upon chin. The pimples had for years been kept under restraint only eliminating all saccharine elements, and fruit from the diet. In e of these cases very great improvement, amounting in three cases t practical cure, was effected by a series of three injections of staphylococcus vaccine.

It may be noted that in none of the above cases was any antise treatment combined with the inoculations. Further, in no case v any restrictions imposed in the matter of diet.

Conclusions.

It is, I think, satisfactorily established by the foregoing cases t chronic staphylococcus invasions, and in particular furunculosis, sycc and acne, can be treated in a very effective manner by inoculations c staphylococcus vaccine. I have elsewhere[1] called attention to broad principles of the therapeutic inoculation of bacterial vaccine the method which is here in question—and to its wide sphere of appli tion. It will therefore here suffice to call to mind that we do not in case of these inoculations supply to the patient protective substan produced in the organism of an animal vicariously inoculated, but induce the chemical machinery of the patient to elaborate by its o efforts the protective secretion which is required for the destruction the invading bacteria. The elaboration of this protective secret proceeds in accordance with the general law that a vaccine introduc into the organism will, given that it is introduced in appropriate do and at proper intervals, call forth a production of the specific bacter tropic substances which are required for the destruction of the bacte against which protection is desired. To comply with the conditions j specified we must employ, as was done in the cases above reported, vaccine of standardized strength. We must, further, for the achiev ment of the best results, measure, before we proceed to reinoculatio ...ced in each case upon the patient's blood by the previo ...ly, it is advisable in obstinate cases to resort to ...he particular strain of micro-organism which h ...grow in the patient's organism. The scienti ...from these observations are reserved for detail ... The reader will, however, note on readi ... improvement invariably went hand in han ...ocytic power, and that the negative pha ...pon inoculation revealed itself to bloo ...ution of the phagocytic power and ... the development of fresh pimpl

[1] ... *British Medical Journa*

boils. This transient and, from the clinical point of view, insignificant ravation of the symptoms is full of instruction, inasmuch as it is icative of the possibility of aggravating the patient's condition by the ployment of excessive and too frequently repeated doses of vaccine.

The Inoculation Treatment of Tuberculosis.[1]

By A. E. Wright.

A Lecture delivered at St. Mary's Hospital to the party of Fre Physicians and Surgeons visiting the London Hospitals.

Method of Immunisation copies Nature's Method of Combating Bacterial Infecti —Definition of the Technical Terms employed in Connexion with Immunisat —Considerations of the Effects which are Exerted upon the Blood by the I culation of Vaccines—Negative and Positive Phase and Cumulation Effect Regulation of Dosage of Vaccines—Antitubercle Vaccines—Koch's Met of Measuring the Tubercle Agglutinins with a View to the Control of Tub culin Inoculations—Discovery of the Opsonins and Exploitation of these the Regulation of the Inoculations—Examples of Desperate Cases of Tubercu Infection treated by Inoculations of T.R. Tuberculin controlled by the Opso Index—Concluding Remarks.

Gentlemen,—There are only two methods by which we may attem to kill off bacteria in the interior of the organism. We may attem to kill them off by introducing antiseptics into the organism ; or we m try to kill them off by the agency of protective substances produc by the organism. The former method has had its day. It has be tried upon the most extensive scale and has failed. It has in particul failed in tuberculosis. I propose to consider with you the alternati method of treating bacterial infection—the method of immunisation.

I will ask you to note on the very threshold that the method immunisation is Nature's method. No one recovers from an acute chronic bacterial disease unless it be by the production of protecti substances in his organism ; no one acquires protection against a disea except, again, by the production of protective substances ; and finall no one lives in the presence of infection and repels that infection excep by the aid of the protective substances of his blood.

It is of the utmost importance that it should come home to yo that we are dealing here, not with mere speculation, but with a genera ization which rests upon a large body of verifiable fact. By the ai of a comparatively simple technique—technique which I have fro time to time described in the scientific journals—it is now possibl not only to demonstrate in a drop of blood drawn from the finger th

[1] Reprinted from the *Clinical Journal*, Nov. 9, 1904.

tective substances which come into consideration in connexion with our ordinary bacterial diseases, but also to measure the content of blood in these substances in an accurate manner. By the help of se methods every laboratory worker will, I think, be able to satisfy nself (*a*) that the blood of those who become the subjects of a bacterial asion is deficient in protective substances; and (*b*) that by an injection corresponding bacterial vaccines the content of the blood in pro- tive substances can in practically all cases be increased. I propose point out to you that this can be done in the case of those who are eady the subjects of tubercular infection.

Let me at the outset define for you the technical terms which I opose to employ, and them briefly rehearse the first principles of the ysiology of immunisation.

Protective substances may be defined as substances which enter o destructive chemical combination with bacteria, or, as the case y be, with other foreign elements introduced into the organism, her directly into the blood-stream, or by hypodermic injection. Such otective elements are never absent from the blood. We can, for tance, in the case of every sample of human blood, demonstrate the esence in it of protective substances which enter into chemical com- ation with the tubercle bacillus. We may signify the fact that these otective substances turn towards and enter into chemical combination th the tubercle bacillus by denoting them, in conformity with the nvenient system of nomenclature devised by Ehrlich, as *tuberculo- pic* substances.

A *vaccine* is any chemical substance which when introduced into e organism causes there an elaboration of protective substances. I n put it more precisely if you will let me put it into technical language. *vaccine is a substance which induces in the organism an elaboration of cterio-tropic elements; a tubercle vaccine a substance which induces in e organism an elaboration of tuberculo-tropic elements.*

We may now take a further step and inquire in what manner the troduction of a vaccine into the system is interrelated with a new rmation of protective substances in the organism.

The interrelation appears to be this. The bacterial substance inocu- ted—and the bacterial vaccine is always a derivative of the bacterial otoplasm—enters into combination with the bacterio-tropic elements ready present in the organism, and thus withdraws from the organism a rtain quantum of protective substances. Under the stimulus of this eprivation the cells of the organism are stimulated to activity, with e result that the bacterio-tropic substances which have been with- rawn are replaced with usury.

This theoretical conception, whatever may be its value, will serve t any rate to impress upon your minds the sequence of events that an be actually observed to occur. Immediately after the injection

of the vaccine there supervenes, as I showed first, in connexion w anti-typhoid inoculation, and afterwards in connexion with a staphylococcus and anti-tubercular inoculation, a *negative phas* that is to say, a phase in which there is a diminished content of p tective substances in the blood. That negative phase is succeeded a *positive phase* characterized by an increased content of protect substances in the blood. This inflowing wave of protective substan rapidly flows out again, but leaves behind in the blood a more less permanently increased content of protective substances. I ha spoken of this whole sequence of events as the "*law of the ebb and f and reflow and maintained high tide of immunity.*"

All this has reference to the effects of a single inoculation und taken with a dose of vaccine which is sufficient to produce a cert constitutional disturbance. When only a small dose of vaccine is in culated the negative phase may be so fugitive as hardly to appear on t record, but the positive phase will be correspondingly diminished. Wh an unduly large dose of vaccine is inoculated the negative phase is pr longed and much accentuated. The positive phase may in such a ca even make default.

We have here earnest reason for considering the question of do It will be obvious that if we, in the case of a patient who is already t subject of a bacterial invasion, produce by the injection of an excessi dose of a vaccine a prolonged and well-marked negative phase, we ma instead of benefiting the patient, bring about conditions which w enable the bacteria to run riot in his system.

If attention to dose is essential in the case of a single inoculation of bacterial vaccine, much more is it essential where we undertake a seri of successive inoculations. We are in such a case superposing the effe of one inoculation upon the other. Now, consideration will show tha we may obtain, according as we choose our time and our dose wisel or unwisely, either a cumulative effect in the direction of a positive phas or a cumulative effect in the direction of a negative phase. We may in other words, by the agency of two or more successive inoculation raise the patient by successive steps to a higher level of immunity, o as the case may be, bring him down by successive steps to a lower leve We can select the appropriate time and dose with certainty only b examining the blood and measuring its content in protective substance in each case before re-inoculating.

If we omit such measurement and work in the dark, our sequen inoculations may quite well fall upon a negative phase period when th content of the blood in protective substances is still below par. In such a case, negative phase would be superinduced upon negative phase and cumulation would take place in the direction of diminishing the patient's resistance. The dangers which might be associated with such a cumulation in the direction of the negative phase are, I think, more than sufficiently exemplified in the fatal results which have in some cases

rvened upon the inoculation of progressively increasing doses of n's old tuberculin.

ou will, perhaps, think that the chances of cumulation in the direction negative phase are very remote if during the course of inoculations linical symptons are watched. No doubt when you have produced ious negative phase effect the fact will be intimated to you by clinical toms. My point is that that warning will be conveyed to you too late. conveyed to you only after you have lost the advantage gained e foregoing inoculations and undone all the good you have done. esults obtained by Madsen and Jörgensen by a daily repeated inocu- upon animals of quite small daily doses of a bacterial vaccine it very clearly the risks associated with any mechanical scheme oculation. In each case the initiation of a series of inoculations followed by a progressive rise in the protective substances of the . But sooner or later a day arrived in each case when the machin- of immunisation had made its maximum response. When that was arrived at the further inoculations served only to bring the nt of the blood in protective substances down with a run. If I out to you in a graphic manner what was obtained in Madsen and nsen's experiments, you will see that small daily inoculations first a gradually ascending curve which passed up into a steep and succeeding immediately upon this rise an equally steep fall, nating in a more gradual slope. You may conveniently think of s a roof-tree curve. Your human patient, who has been raised to her level of immunity by successive small inoculations, will, in nanner sooner or later, if too much is required of him, cease to nd to your inoculations, and simply slide down the slope of a "roof-' curve. If you have not been measuring the content of his blood otective substances, your patient may probably have arrived at er level of resistance than that from which he originally started symptoms of intoxication draw your attention to the fact.

ese fundamental principles of the art of immunisation having made clear, I will pass on and consider with you the question of the cle vaccine.

ou will remember that I defined a bacterial vaccine as a substance was capable of inducing an elaboration of anti-bacterial sub- s in the organism, and a tubercle vaccine as a substance which was le of inducing an elaboration of anti-tubercular, or, as I prefer them, tuberculo-tropic substances. I indicated to you also that bacterial vaccine is derived from the corresponding bacterial e. Such vaccines need not, as was assumed by Pasteur, consist ing cultures. It has been adequately established in connexion many bacterial vaccines that their vaccinating efficacy is not ed by sterilization by the action of moderate heat (60° C.). I lay stress on the fact that this holds true also in connexion with le vaccine.

We pass to consider in what form a tubercle vaccine is access
It is accessible in the form of Koch's T.R. tuberculin. This consis
may remind you, of a fine tubercle powder (obtained by comminu
tubercle bacilli by the action of machinery) which has been bro
into suspension in definite quantity (10 mgr. to 1 c.c.)[1] in dilute
cerine.

You can, as already indicated, provide for the efficient steriliza
of this vaccine without impairing its efficacy by heating the prepara
to 60° C. for one hour. Most of the inoculations I have underta
have been with a vaccine thus treated. I, however, propose herea
to experiment with a vaccine which I have recently made on exa
the same lines as the staphylococcus and typhoid vaccine—to wi
simple sterilized suspension of tubercle bacilli standardized by the
cedure for enumeration under the microscope which has been descr
by me elsewhere.[2]

I pass from the question of the vaccine to the question of the dos
and in particular to the question of the scheme of operations. In o
to arrive at the scheme of operations to be adopted in the therape
application of the vaccine to those who are the subjects of tuberc
infection, we have to consider the conditions with which we are
fronted. The tubercular patient has probably at the onset, as c
pared with the normal man, possessed a deficient power of resista
Again, he has in most cases not responded to infection by any incre
elaboration of protective substances. This was, at any rate, the
dition of affairs in the seventeen cases[3] of localized tubercular infec
which I have tabulated in the *Royal Society's Proceedings*, vol. lx
October, 1904.[4]

In dealing with these cases the following will appear to be our pr
line of policy. We have by increasing the content of the blood in
tective substances to try to forestall any dissemination of the tuberc
bacilli by the channel of the blood stream. Further, we have to
to inhibit the growth, and if possible to bring about the destructio
the tubercle bacilli in the local nidus of infection by leading thro
that nidus in a continuous stream a lymph rich in protective substanc

I may point out in this last connexion that protective substa
are continuously withdrawn from the lymph as it comes in co
within the infected tissues with the invading bacilli. Captain Do
and I have shown in the papers already referred to that the stag
lymph in an ordinary abscess contains hardly a trace of antibact

[1] But *vide supra*, p. 123, *note* 2. [2] *Lancet*, July 5, 1902.

[3] There is, it is to be noted, also another class of cases where the phenon
are apparently complicated by processes of self-immunisation comparable to
found in connexion with general infections. This class of cases, which num
among it many of the subjects of chronic phthisis, is here provisionally left o
consideration.

[4] Pp. 118 *supra*.

stances, and that the evacuation of the contents of an abscess and the lication of fomentations effect a douching of infected tissues by a ph whose content in protective substances is not less than that of circulating blood. Further, it has been shown by us in a case of ercular peritonitis that the fluid contained in the peritoneum was times poorer in protective substances than the patient's blood. In nexion with this, we put forward the suggestion that the evacuation he old and stagnant lymph and the transudation of new and potent ph from the blood vessels furnished the probable explanation of the antage which is so often obtained in tubercular peritonitis from the cuation of the ascitic fluid.

In the scheme of operations sketched out above we must count *as lost* to the patient any period during which the protective substances d in his blood at the low level at which those substances are wont to d in the untreated patient.

We must count as *a period of retrogression* for the patient any period ing which his protective substances sink below the ordinary low level is untreated state.

We must count as *a period of progress* for the patient every day during ch the content of his blood in protective substances stands at a er level than that of his untreated state.

Lastly, we must *deduct from the period of progress* every hour, n, by reason of the stagnation of the lymph circulation in the seat of ction, the protective substances of the blood cease to come into lication upon the tubercle bacilli in the foci of infection.

Only those who will meditate upon these points will appreciate to the the difficulties which confront the immunisator.

I hope to be able to convince you that, these multiform difficulties withstanding, it has been possible in nearly every case to gain some antage from the injection of the tubercle vaccine, and in some ningly desperate cases to achieve what will, I think, appeal to you as nplete success. I think the cases I propose to show you warrant in hoping for even better results in the future, when the programme ch I have sketched out shall have been more closely adhered to n it was possible to do in connexion with these first tentative efforts.

Before I deal with the cases I would turn aside for a moment and sider with you what has emerged with respect to the protective sub- nces of the blood.

When I addressed myself, two years ago, to the study of the pro- tive substances which come into consideration in connexion with the ercle bacillus, nothing was known about these beyond the fact that it possible by the aid of the so-called homogeneous tubercle cultures Arloing, and also by the aid of homogeneous suspensions of a tubercle vder (made by Koch by the trituration of tubercle bacilli) to obtain ertain instances—in particular, as Koch showed, in the case of patients had been treated with his T.R. tuberculin—an agglutination reac-

tion comparable to that obtained in connexion with typhoid fever
antityphoid inoculation.

A fallacy in the form of spontaneous agglutination was associ
with the technique prescribed by Koch. It, however, emerged in
course of my work that this fallacy could be avoided by employing
lieu of the physiological salt solution used by Koch for the suspen
of his powder, a salt solution ten times weaker. I may, perhaps, cl
that by attention to this point, and by the adoption of the system
technique described by me in the *Lancet* of July 25, 1903, the difficu
associated with the measurement of the tubercle agglutinins in the bl
have been overcome.

Until some months ago the measurement of the content of the bl
in these tubercle agglutinins constituted the only means of obtain
information from the patient's blood with regard to the success or
success of the attempted immunisation. The investigation of the bl
by that method gave all too dim a light.

Of late, working on the problem as to how the organism prot
itself against those numerous species of pathogenetic mic
organisms which offer absolute resistance to the bactericidal act
of the serum, and in particular on the problem as to how the hum
organism protects itself against tubercle and staphylococcus infectio
and as to how it reacts to the inoculation of tubercle vaccine and stap
lococcus vaccine, I have found, in conjunction with Captain Stew
Douglas, that there exists in the normal serum, and there ex
in larger quantity in the serum of the successfully inocula
patient, an element which enters into chemical combination with
staphylococcus, the tubercle bacillus, or other micro-organism, in such
manner as to prepare it for phagocytosis. We have called that p
tective element an *opsōnin* (Latin, *opsōno*—I cook for table, I prep
pabulum for). We have demonstrated that phagocytosis cann
take place apart from the action exerted by the specific opsonin up
the micro-organism, and we have shown that opsonic action is destroy
by heating the serum to 60° C. In this opsonin we have, it wou
seem, an essentially important protective substance. It is, further,
substance which lends itself to very accurate measurement.

That measurement is effected by a modification of Leishman's metho
i.e., by mixing together in a capillary tube in each case one volume of t
patient's serum, one volume of a suspension of the tubercle bacillus, a
one volume of washed corpuscles obtained from the citrated blood
a normal man. The capillary tube is now placed in an incubator f
any convenient period, generally for fifteen to twenty minutes. Aft
that period microscopic films are prepared. After appropriate staini
the first thirty to forty white corpuscles which come into view are e
amined, the number of bacilli ingested by each white corpuscle bei
noted down. The "phagocytic count" is then arrived at by addi
together the number of ingested bacteria and dividing by the numb

leucocytes examined. This "phagocytic count" is compared with
" "phagocytic count" obtained in films made with the contents of a
oillary tube similar in all respects, except in the respect that there has
en employed in it, instead of the patient's serum, the serum of a normal
rson. The ratio in which the phagocytic count of the patient's blood
nds to the phagocytic count of the normal blood (taken in each case
unity) is conveniently spoken of as the "opsonic index." By the aid
this method the patient's progress or regress can be very accurately
lowed.

I have left myself only too little time to go into the history of the
tients whom I have here for your inspection, and to supplement what
can show you here by reference to other cases.

CASE 1.—The woman you see before you has, as you see, the aspect of
oust health. Her history is as follows: Early in January, 1903, her
e being then thirty-three—she was admitted to Mr. Silcock's ward for
dominal pain and distension associated with fever and loss of weight.
ese symptoms had been first noticed in the previous September. Mr.
cock operated on January 22, the abdomen being opened by an incision
e inches in length. The typical appearances of tubercular peritonitis were
ought into view and the surface of the intestine was seen to be in
ces studded with miliary tubercles. After the evacuation of a very
nsiderable quantity of fluid the peritoneum was washed out and the
und was closed, a drainage-tube being left in position.

After the operation the fever still continued. It reached 102° F. every
ening during the first week; it reached 101° F. every evening for the
xt fortnight; and it still ranged up to 100° F. every evening two months
ter the operation. All this time the wound was continuing to dis-
arge, and the patient was becoming very weak and emaciated—being
ite unable to turn unassisted in bed.

Treatment by tuberculin inoculation was begun on March 17th.
ithin a few days the evening temperature had sunk away to 99°, and
came down to the normal on April 28, and remained normal (except
hen slightly disturbed by certain of the tuberculin inoculations) for
e three months which the patient still spent in hospital. From the
ginning of the tuberculin treatment onwards the patient improved
strength and put on flesh. In June she was able to sit up in the
ternoons. Her body weight was now 105 lb. In July she was dis-
arged from hospital, the abdominal wound having now completely
ealed except for a narrow sinus. The tuberculin treatment was con-
nued, the patient being treated first at home and afterwards as an out-
tient.

Six months afterwards the sinus had completely closed and the
tient's weight had by March, 1904, increased to 132 lb. She had,
other words, increased 27 lb in weight in six months, and had passed
thin a year from a seemingly desperate condition to a condition of,
think, perfect health, such as you now see her in.

CASE 2.—The patient whom I now present to you is, as you see, a v
well nourished woman, aged forty-three years, who, perhaps, looks a li
pale. She was admitted to the hospital under the care of Dr. Lee
the middle of March, 1903, complaining of very frequent micturit
associated with severe local pain on micturition and dragging pain in
loins, in particular on the left side. The urine contained pus, epithe
casts, and tubercle bacilli in such numbers that they could be dem
strated in large clumps in every field of a microscope in preparati
prepared from the urinary sediment. Examination of the blad
revealed the existence of a large open ulcer. The kidneys were enlar
and tender, the left one in particular was affected, and suspicious si
were detected in the apex of one lung. Tuberculin treatment was beg
in the middle of April. The effect exerted upon the body weight dur
the period the patient was in hospital is exhibited in the column of figu
which I have placed on the board :—

	lb.		lb.
April 20	91	June 15	103
„ 28	96	„ 22	105
May 11	92½	„ 29	107
„ 19	93	July 6	109
June 1	96	„ 13	107
„ 8	101		

To one incidental feature in that record I should like to draw yo
attention. The drop of body weight which is recorded on May
coincided on the one hand with the development of increased local pa
and symptoms of giddiness and flushing, and on the other hand w
a rapid fall in the agglutinating power of the blood, which is displayed
the agglutination chart[1] I have placed on the table. These were a
I take it, symptoms of the supervention of a negative phase depende
upon a too hasty inoculation of progressively increasing doses of vacci

After leaving the hospital in July, much alleviated in the matt
of pain and frequency, the patient attended as an out-patient, and und
the treatment her weight in September, 1903, reached 119 lb. T
tuberculin inoculations have been continued up to recently. All th
while the tubercle bacilli, which have been examined for almost eve
ten days, have become gradually less numerous. Since May last they ha
completely disappeared from the urine. The patient, none the less, st
suffers from serious bladder trouble—due, as appeared recently o
examination, to cicatrization and great thickening of the bladder wal
and possibly to some super-added ulceration referable to septic invasio
by the bacillus coli and by a Gram-staining diplococcus—micro-orga
isms which have been throughout present in millions in her urine.

You will see from her good general condition and from the histor
of the facts I have given to you that the tubercular infection has in a
probability been overcome.

[1] *This chart is reproduced on* *p.* 271 *infra.*

Case 3.—The patient I now present to you is, as you see, a man of bout thirty years of age. His history is as follows: In the autumn of 902 the patient developed tubercular glands on the left side of the neck nd a tubercular abscess on the point of the left shoulder of the same side. Ie was admitted to this hospital, and was operated upon for the first ime in January, 1903. The wound becoming invaded with tubercle nd refusing to heal. further operative procedures were undertaken. n all six successive scraping, extirpating and skin-grafting operations vere undertaken during the course of the year, the wound becoming in ach case re-infected, and the area of ulceration being increased. In December, 1903, when the patient came up for treatment by inoculation, he whole area from the point of the shoulder to the base of the ear ormed a single deep eroded ulcer. The pinna of the ear was alf eaten away (you can still judge of that), and immediately under-eath it there was a deep crateriform ulcer which looked as if it was oing to break into the pharynx. The left side of the face was dis-orted by swelling, giving the patient the appearance which would go long with a one-sided attack of mumps. The axilla was occupied by a land which was as large as a pigeon's egg, and the patient was haggard nd very emaciated. As you see for yourselves, he is now, if somewhat allow, yet a not unhealthy-looking man. You will see, as he strips, hat the whole of the area I have described to you as previously occupied y the tuberculous ulcer, with the exception of an area about the size f a threepenny piece, which is still covered by a scab, has cicatrized nd skinned over. You see how soft and elastic that skin is. It differs rom the rest of his skin only in being a little pinker. The glands in the xilla can now no longer be felt.

I may mention that, in addition to inoculating this patient with uberculin, I gave him one or two inoculations of a staphylococcus vaccine vith a view to purging the ulcer from the staphylococcus which had nvaded its surface. Further, I would point out that I have during the ast six weeks or two months succeeded in hurrying up the final processes f repair by painting upon the wound a 20 per cent. solution of gelatine, o which I added 2 per cent. of formalin. This sets into firm, insoluble ntiseptic skin.

In connexion with this case, I may perhaps refer to two further ases of tubercular glands to which I have applied the inoculation reatment.

In the first of these cases the patient was a young married woman who had undergone at the hands of two distinguished London surgeons hree successive operations for the extirpation of glands. When she resented herself for treatment in January last three or four glands ould be felt in the neck, the largest one being about the size of a small walnut. After the diagnosis of tubercle had been confirmed by a test noculation undertaken with Koch's old tuberculin, the vaccinal treat-ment with the T.R. tuberculin was inaugurated. After six inoculations

conducted with doses which were gradually increased from $\frac{1}{500}$ mgr. a maximum of $\frac{1}{50}$ mgr.,[1] the glands could no longer be felt and the dr ging pains in the neck had entirely disappeared. A period of th months was occupied by these inoculations. Since the date of disappearance of the glands a few more reinforcing inoculations have b given with a view to preventing any return of the symptoms. Up the presence there has been no recurrence.

The second of the cases of tuberculous glands above referred to the case of the wife of a medical man. She had suffered from chi hood from swollen glands in the neck on one side of the neck. Th had become the source of constant dragging pains, and the largest gla situated under the angle of the jaw, was large enough to produce so disfigurement. In this case also the glands have been very considera reduced in size, while the dragging pains have completely disappear and the whole physical condition has improved in a remarkable mann

I ought, perhaps, here to point out that if I include in my catego of tubercular glands treated two cases of Hodgkin's disease, which we both in a very grave condition when they were taken in hand, I have addition to the three cases already recounted also two failures to reco In both these cases the diagnosis of tubercular infection was confirmed a test inoculation of Koch's old tuberculin. In both cases very small dos of T.R. tuberculin (doses of $\frac{1}{500}$ to $\frac{1}{100}$ mgr.[1]) were employed. In ea case very severe constitutional symptoms followed upon every inoculatio and again in each case only a small and transient production of protecti substances (only the tubercle agglutinins were measured) was achieve

Case 4.—The next case I present to you is a woman with lupus. S has, as you see, lost her left arm, and you can see that the stump of th arm, the sternum, the right hand, and the face and neck on both sides a extensively affected with the disease.

Her history is as follows: She developed a tubercular infection of t glands of the neck at the age of fourteen. Then suppuration supervene and the abscesses were opened, the wounds became infected and oth glands also became involved. Later tubercular disease develope in the little finger of the right hand. The two terminal joints of tha finger were removed fifteen years ago, when the patient was sixtee About this time lupus broke out on her face and on her left arm and han At the age of nineteen the patient underwent treatment with Koch original tuberculin. She received three to four inoculations a day (th total of her inoculations amounting to 150). This treatment resulte in violent inflammatory reaction in the patches of lupus, a piece of bon sloughed out of her left arm, and she remained in hospital seriously il for thirteen weeks. She attributes—and no doubt rightly—the aggra vation of symptoms and ultimate loss of her arm to these inoculations

We can see that there must have been produced a cumulativ negative phase.

[1] *Vide* *p.* 113, *note* 2.

After a respite vigorous treatment was resumed in another hospital. The lupus patches were then frequently scraped and many glands were extirpated from the neck.

In 1900 the Finsen light treatment was resorted to and was persevered in for eighteen months. This effected superficial improvement in the condition of the face and neck, but the disease continued to spread in the deeper structures, and in particular in the bones of the left arm. Finally it became necessary to amputate this limb.

The disease now re-invaded the stump and broke out in the point of the shoulder and in the front of the chest.

Röntgen rays were now tried, unavailingly. Finally, in December, 1903, the patient, who was then in a very reduced physical condition, was referred to me by Dr. Graham Little for treatment by tuberculin inoculations. You see now that the patient is in a tolerably satisfactory condition in the matter of her general health. Her body weight has gone up and has reached 141½ lb., as much as 5 lb. having on one occasion been gained in the interval between two successive inoculations. The discharge from the sinus over the sternum has practically ceased. The same holds true of the sinus in the stump of the left arm. The open sore on the point of the shoulder has healed up, and the patches of lupus on the face seem to me to be becoming quiescent.

Case 5.—The patient I now present to you is, as you see, another case of lupus. She has the appearance of a child, but is, she tells me, in her twentieth year. When referred to me for treatment by Dr. Graham Little she was extremely emaciated, and no doubt under-fed, her bones protruded through the skin of her back somewhat after the fashion of the bones in fish that has been split and dried. The point of her nose, which you now see presents the appearance of healing, was covered with a thick mass of scabs superposed upon a very angry-looking patch of lupus. The angle of the jaw and the front of the neck were occupied by patches of lupus in a similar condition. These are, as you see, now represented by somewhat swollen cicatrices. Both her feet and her hands were affected with lupus. Her hands in particular constituted a mass of ulceration, the bones of the hand being also affected in many places. The condition of these is somewhat ameliorated.

I have under treatment, or have had under treatment, also other cases of lupus for different periods. I think I can say that every one of them has improved, with the exception of one of the first cases I treated. Here, owing, I think, to the too rapid increase of doses of vaccine, the protective substances (agglutinins) which had developed under the stimulus of the first inoculations were subsequently lost.

And now just a few words in conclusion. I have endeavoured to place before you the principles which ought, I think, to guide us in the therapeutic inoculation of bacterial vaccines. I have dealt more particularly with the results of the application of that method to the treatment of tuberculosis. But I would point out to you that the method

is one which has a perfectly general application in connexion wi localized bacterial infections.

I have here, for instance, an example of the application of the meth to the treatment of a chronic staphylococcus infection. The patie I here show you, one of our medical students, is one out of a series fifty or more patients which I have treated (almost all with comple success) by inoculations of staphylococcus vaccine for different forms staphylococcus infection. You have here, as you see, to deal with a ve severe case of acne. Before inoculation each acne spot became, as usually does, the seat of a staphylococcus infection and was convert into a pustule. The inoculations, which were undertaken about a ye after, have accomplished their purpose. The patient is, as you se free from every trace of pustulation. Time does not allow even of m outlining the many other practical applications which can be made of t method of therapeutic inoculation with sterilized bacterial culture It may, however, interest you to hear that the method has alread been successfully applied by me to widely different cases. I may pa ticularize : (1) a case of acute coli infection of the biliary passages, whe after removal by operation of an impacting biliary calculus, the fev and jaundice continued, and the bile was flowing away through th external wound by reason, as it seemed, of the plugging of the bile-duc by inspissated mucus ; (2) a case of coli cystitis which had continue for sixteen years : (3) a case of a localized infection by the micrococcu Melitensis supervening upon an attack of Malta fever ; and lastly (4) case of pneumococcus infection of the salivary glands which was as sociated with very burdensome salivation. In this last case, in contras to the others, amelioration only has been achieved.

On the General Principles of the Therapeutic Inoculation of Bacterial Vaccines as applied to the Treatment of Tuberculous Infection.[1]

By A. E. Wright.

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Preliminary Matter—Train of Events which follows upon the Inoculation of a Bacterial Vaccine—Train of Events which follows upon the Inoculation of a Series of Doses of a Bacterial Vaccine—Consideration of the Principles which ought to Regulate the Dose of Vaccine—Manner in which the Organism conducts itself when it becomes the Subject of Bacterial Invasion; and Discrimination of Bacterial Infections into (*a*) Bacterial Infections where the Machinery of Immunisation is Inactive, and (*b*) Bacterial Infections where the Machinery of Immunisation is called into Action—Conditions under which Pathogenetic Micro-Organisms cultivate themselves in the Interior of an Infected Organism—Treatment of Strictly Localized Tubercular Infections by the Aid of Therapeutic Inoculations of a Tubercle Vaccine—Discussion of the Means which are available for sending a Stream of Antibacterial Lymph through the Focus of Infection—Digression on the Results of Ordinary Surgical Methods as Applied to the Treatment of strictly Localized Tuberculosis—On the Results which have been obtained by the Treatment of Localized Tubercular Infections by the Aid of Therapeutic Inoculations of a Tubercle Vaccine (Koch's New Tuberculin) controlled by Determinations of the Opsonic Index—Treatment of Systemic Tubercular Infections by the Therapeutic Inoculation of a Tubercle Vaccine—Conditions which we have to take into Account in Connexion with Ordinary Continued Fevers—Special Conditions which we have to take into Account in Connexion with Pyrexial Phthisis and other Localized Tubercular Infections which are associated with Pyrexia—Possibility in Connexion with Pyrexial Phthisis and other Localized Tubercular Infections which are Associated with Pyrexia of quieting the Circulation and Staunching the Lymph Stream in such a way as to arrest the Auto-Inoculations, converting the Systemic Infection in this manner into a purely Localized Infection—Consideration of the Question as to how far the Patient has been brought in the Direction of a Cure when his Pyrexia has been abolished and his Auto-Inoculations have been arrested by Confinement to Bed—Programme of Treatment which would appear to be Indicated in the Case of Pyrexial Phthisis, or other Localized Tubercular Infection which may be associated with Pyrexia.

1 Reprinted from the *Transactions of the Medico-Chirurgical Society*, vol. lxxxix, 1906.

PART I.

PRELIMINARY MATTER.

What I have to say to-night on the subject of the treatment of tuber culous infection by the therapeutic inoculation of tubercle vaccine ma conveniently be prefaced (*a*) by a recital of the train of events whic supervene in the blood upon the inoculation of a bacterial vaccine, or, as the case may be, upon a succession of such inoculations; (*b*) by consideration of the principles which may properly guide us in determinin in the case of each successive inoculation the dose of vaccine to b administered; (*c*) by a brief account of the manner in which the organism conducts itself when it becomes the victim of a bacterial invasion; anc (*d*) by an exposition of the conditions—so far as these are known to us— under which pathogenetic bacteria cultivate themselves in the infected organism.

Train of Events which follows upon the Inoculation of a Bacterial Vaccine.

The changes in the antibacterial power of the blood which supervene upon the inoculation of a bacterial vaccine were for the first time investigated by the aid of quantitative methods and upon man in connexion with my work on antityphoid inoculation. That work has been followed up by similar researches conducted by myself and my pupils and fellow-workers in connexion with the inoculation of Malta fever vaccine, tubercle vaccine, plague vaccine, pneumococcus vaccine, staphylococcus vaccine, streptococcus vaccine, gonococcus vaccine, proteus vaccine, and a series of vaccines made from different strains of the *Bacillus coli.* All of these vaccines, with the exception only of the plague vaccine, have come into application in connexion with the treatment of the corresponding bacterial infections.

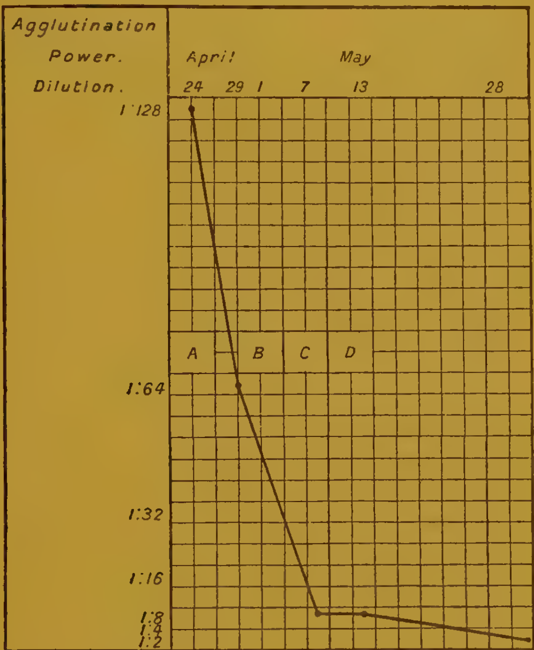
Upon the inoculation of each of these vaccines without exception there has followed one and the same train of events. That train of events is as follows: (1) Upon the inoculation of the vaccine there supervenes a period of intoxication which is characterized by a decline in the antibacterial[2] power of the blood. This "negative phase" is more or less accentuated and prolonged, according as a larger or smaller dose of the vaccine is inoculated. In the former case the negative phase may disclose itself to clinical observation by a temperature reaction and con-

[1] The term "vaccine" is here and throughout this paper employed to denote a sterilized and standardized suspension of micro-organisms.

[2] The particular antibacterial element which was measured was in the large majority of cases the *opsonin.*

tutional disturbance. In the latter case the negative phase may quite unaccompanied by clinical symptoms. (2) Upon the negative ase there follows a "positive phase." This phase, whose char-eristic feature is an increase in the antibacterial power of the blood, responds to a period of increased resistance. The curve whose trace

CHART 1 (by Author).



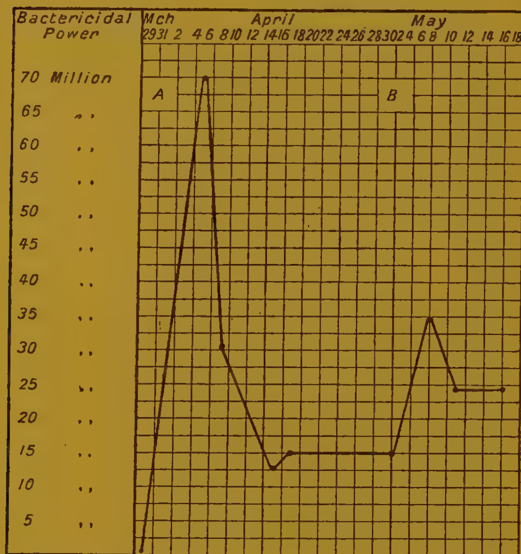
lating to E. S——, a case of tubercular cystitis, treated by inoculations of new tuberculin (Case 4, p. 292 *infra*), showing that a cumulation in the direction of the negative phase is produced by the inoculation of a series of inappropriately adjusted and inappropriately interspaced doses of a bacterial vaccine. A. Inoculation of 0·01[1] milligramme of the new tuberculin. B. Inoculation of 0·025 milligramme of the new tuberculin. C. Inoculation of 0·05 milligramme of the new tuberculin. D. Inoculation of 0·2 milligramme of the new tuberculin. The method employed for testing the blood was that described by the author, *Lancet*, July 23, 1903.

s forth the changes in the anti-bacterial power of the blood runs up in ny cases into a sharp peak and sinks away, first comparatively rapidly d afterwards more slowly. There is associated in many cases with the max of the positive phase a sense of increased physical vigour and a ry pronounced feeling of well being. (3) After the negative and positive ase, which train of events I have ventured to speak as of "the ebb and

[1] See p. 123, *note* 2.

flow and reflow of the tide of immunity," the blood may be maintain for a variable period (after tubercle inoculations, when *the infection been satisfactorily got under*, occasionally for as long as a month) a somewhat higher level of antibacterial power than before inoculati Or—and this in connexion with inoculations with tubercle vaccine i

CHART 2 (by Author in conjunction with Captain W. Glen Liston, I.M.S.).



Relating to a rabbit which was being immunised against the typhoid bacill showing that a cumulation in the direction of the positive phase may be obtain by the inoculation of appropriately adjusted and interspaced doses of a bacter vaccine. A. First inoculation ; 5 cubic centimetres broth culture of the typh bacillus. B. Second inoculation ; 5 cubic centimetres broth culture of t typhoid bacillus. The bactericidal power which is charted represents t bactericidal power of, in each case, 1 c.c. of freshly drawn serum. The meth employed for measuring the bactericidal power of the blood was that describ by the author, *Proc. Roy. Soc.*, vol. lxxi, 1902.

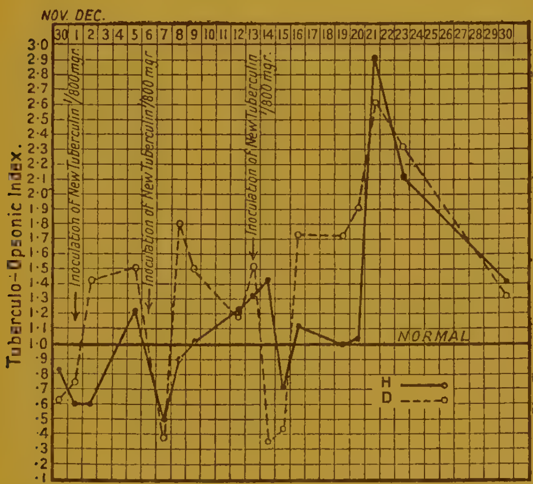
more usual event—the antibacterial power of the blood may over a over again fall back after ten days or a fortnight to the level at whi it stood anterior to inoculation.

Train of Events which follows upon the Inoculation of a Series of Do of a Bacterial Vaccine.

I originally pictured to myself that a cumulative effect in the directi of the negative phase such as is exhibited in the curve here shown (Cha

would occur in a regular manner where re-inoculation is undertaken the negative phase of a preceding inoculation, and that vice versâ a nulative effect in the direction of the positive phase such as is exhibited the companion diagram here shown (Chart 2) would be achieved in a ular manner by reinoculating in the positive phase of a previous

Chart 3 (by Author).



ating to H and D, two children with tuberculous glands, who were treated with therapeutic inoculations of new tuberculin. The curve shows the condition of the blood in each case before inoculation, and the changes in the tuberculo-opsonic power which supervened upon the three first inoculations.

culation. Further experience has shown me that, while cumulation the direction of the negative phase is a phenomenon which must every-ere be reckoned with, it is in connexion with inoculations undertaken h tubercle vaccine difficult, if not impossible, to achieve cumulation the direction of the positive phase. This is clearly brought out in nnexion with the two traces in Chart 3, which show the result of an deavour to achieve in connexion with the inoculation of tubercle ccine a cumulative effect in the direction of the positive phase. In w of this, and a number of similarly unsuccessful endeavours, ave, in connexion with the inoculation of tubercle vaccine, put out of thoughts all idea of cumulating positive phase on positive phase. I now content to treat each inoculation as an independent event, ulating my dose as described.

Consideration of the Principles which ought to Regulate the Dose of Vaccine.

There appears to be everywhere a fixed idea that to secure the greatest yield of protective substances we ought in each case to begin with a dose which produces a certain amount of constitutional disturbance, and that we ought in subsequent inoculations to employ doses which increase by geometrical progression. This fixed idea rests as a matter of fact upon the preconception that immunisation cannot be either initiated or followed up apart from constitutional disturbance, and on the further preconceptions that the capacity of the organism for immunising response is practically unlimited, and that the yield of antibacterial substances will increase *pari passu* with the dose. This is not so. I obtain almost every day maximal immunising responses from the inoculation of doses of tuberculin which have not produced any constitutional disturbance. Further, I have for periods extending over a year continued to inoculate with doses of new tuberculin corresponding to from $\frac{1}{1000}$ to $\frac{1}{600}$ milligramme of tubercle powder [1] without registering any falling off in the immunising response. Again, I have in some of these cases repeatedly registered worse and not better results whenever larger doses than these were employed. Lastly, I have before my mind the fact that the horses which are, in connexion with the manufacture of diphtheria antitoxin, inoculated with large doses of diphtheria toxin, all sooner or later lose their power of responding to the stimulus of inoculation, and recover that power of response only after a long period of rest.

In view of these facts I would submit that the whole question of dosage requires to be reconsidered. For myself, I am day by day more impressed with the fact that the machinery of immunisation can be brought into action by very small stimuli, and that it can very easily be overtaxed. In accordance with these facts I regard it as a matter of great moment, especially in connexion with immunisation against tubercle, to employ in every case the smallest doses which will elicit a satisfactory response; to repeat the dose only when the effect of the preceding inoculation is passing off; and to increase the dose only when it becomes clear that the dose previously employed is ceasing to evoke a sufficient immunising response. Acting in accordance with this principle, I now begin with a quantum of tuberculin corresponding to no more than $\frac{1}{1000}$ milligramme of the tubercle powder, and now never advance to doses larger than $\frac{1}{600}$ milligramme.[1]

I may before passing on just refer to two further points with regard to the dosage of tubercle vaccine.

Where on observing the results of a series of inoculations I find that the negative phase phenomena are becoming with each inocu-

[1] The doses in this paper have reference in each case to the weight of tubercle powder stated to be held in suspension in the new tuberculin as issued. (*But vide* *p.* 123, *note* 2.)

ion more pronounced, I know that I am exceeding my proper dose. here, on the contrary, the negative phase phenomena are becoming er each inoculation less well marked I know that I am employing the oper dose and am making good progress.

The last point to which I would call attention is this: Where a dose has en administered prematurely, or where too large a dose has been adminis-ed, there may result from this, in the case where the positive phase of previous inoculation has not yet exhausted itself, only the cutting ort of that phase, or, as the case may be, the production of a negative ase which is unduly accentuated and which is followed up somewhat rdily by a positive phase. But the case will also occur where, after administration of an excessive dose or premature reinoculation, the sitive phase makes default. Where the positive phase is long delayed ake it that the proper policy is not to wait indefinitely for its arrival t to reinoculate again with a smaller dose as soon as ever the blood sturbance has come to rest.

anner in which the Organism conducts itself when it becomes the Subject of Bacterial Invasion; and Discrimination of Bacterial Infections into (*a*) Bacterial Infections where the Machinery of Immunisation is Inactive, and (*b*) Bacterial Infections where the Machinery of Immunisation is called into Action.

Consideration will make it clear that a knowledge of the effects exerted on the blood by inoculations of bacterial vaccines will not, taken by elf, constitute a sufficient equipment for the physician who desires come actively to the aid of the organism when invaded by pathogenetic cteria. It will manifestly be quite out of question for us to assist in intelligent manner by inoculation until we have ascertained what tion, if any, the infected organism is taking with respect to the invading crobes.

While we are only upon the very threshold of knowledge with respect these subject matters, certain of the broad general principles have eady emerged; and these, inasmuch as they seem to be of absolutely ndamental importance in connexion with the treatment of bacterial sease, I will venture to lay before you. It emerges in a very clear anner from the already very many thousands of quantitative estimations of the opsonic power of the blood which I and my fellow-workers ve conducted in connexion with many forms of bacterial disease, that cterial infections distribute themselves naturally into two categories. one class of infections the opsonic power with respect to the infecting cro-organisms hardly varies from day to day, remaining always ferior to that of the normal blood. In another class of infections the sonic power is continually fluctuating—the range of variation being om far below the normal to far above the normal. These two categories infections correspond respectively to *strictly localized* and *systemic fections.*

An explanation of the different findings in these two classes of ca readily suggests itself. We are, I think, warranted in conceiving of t low opsonic power which is found in association with strictly localiz infections as a condition which dates back to a period anterior to infectic Further, we are, I think, warranted in attributing the circumstance th the opsonic power of the blood remains, in the case of strictly localiz bacterial infections, persistently low, to the default of those immunisi stimuli which are supplied by the entrance of bacterial elements into t blood. And again we are, I think, warranted in conceiving of the fluctu tion of the opsonic power between high and low, which is found in asso ation with systemic infections, as the expression of a periodic activati and inhibition of the machinery of immunisation, brought about by t conveyance of bacterial elements into the blood, in appropriately or, the case may be, inappropriately adjusted and interspaced doses.

Our strictly localized, and our systemic bacterial infections wou in this manner resolve themselves into a category of infections whe the stimuli which call forth an increased elaboration of protective su stances make default; and into a category of infections where we ha to reckon with the delivery of, oftentimes ill-adjusted and oftentim inappropriately interspaced, auto-inoculations.

In association with this difference between infections which evol immunising responses and infections which evoke no such responses the emergies a distinction which is of absolutely fundamental importance.

Systemic infections—provided always that the machinery of immun sation is not overtaxed—are infections which terminate ordinarily death, or in a cure—that cure when it occurs being never indefinite delayed.

Strictly localized infections do not tend to get well. They are cha acterized by an altogether indefinite duration.

I need not remind you that, while an acute specific fever will ord narily run its course within a limit of one, two, or three weeks, a strict localized infection, such as lupus, may commence in earliest infancy an run on through sixty or even more years, terminating only with the li of the patient.

Conditions under which Pathogenetic Micro-Organisms cultivate them selves in the Interior of an Infected Organism.

With the discovery of the bactericidal properties of the blood c susceptible animals the problem presented itself as to how bacteria coul maintain their existence in an organism which was furnished with thes bactericidal elements. With the discovery of the agglutinating powe of the blood, the parallel problem as to how the infecting micro-organism can remain unagglutinated in the interior of the organism; an with the discovery of the opsonic power of the blood, the similar probler

to how the infecting micro-organisms can escape phagocytosis in the ganism in the presence of leucocytes, presented themselves for solution.

The general problem as to how bacteria can maintain themselves an organism which is provided with antibacterial substances has been alt with by Metchnikoff by a procedure similar to that which was opted by Alexander the Great in the case of the Gordian knot. Metchniff's method of dealing with the problem is to contend that bactericidal, glutinating, and antibacterial elements generally make their appearce in the blood only after this has been withdrawn from the blood vessels phagocytes have dissolved in the blood fluids. This contention—iich is so congenial to every one who desires to leave out of his reckongs in dealing with bacterial diseases everything that relates to the tibacterial power of the blood—seems to me to be in conflict with the iole of the experience which is won by a systematic comparison of the nical condition of the patient with the result of quantitative measureents of the antibacterial power of his blood.

I will ask you, therefore, to consider with me whether we have t a very simple solution of the problem before us in the consideration at the infecting micro-organisms cultivate themselves in the organism every case in regions of *lowered bacteriotropic pressure*—i.e. in regions iere antibacterial substances are absent from the tissue fluids or where ese contain antibacterial substances in diminished quantity as compared ith the circulating blood.

This theory, which was first enunciated by me in a paper written in njunction with Lamb on "The Distribution of the Agglutinins in the rganism in the Case of Typhoid and Malta Fever," [1] was shown by us furnish a key to the explanation of our findings in connexion with ose infections. Later this theory was shown by Lamb [2] to furnish e key to his findings in connexion with the bacteriolysins in spirillum ver. Lastly, in conjunction with Douglas,[3] and afterwards with Reid,[4] have been able to show that this theory furnishes the key to the disibution of opsonins in the infected organism, in the case of tuberculosis id a large number of other bacterial infections.

Let me try to put the situation before you as I conceive of it in conexion with micro-organisms cultivating themselves in the tissues. I nceive that these are cultivating themselves under conditions which not even remotely resemble those which they would have to confront the circulating blood. In the case of bacteria in the actual bloodream all the anti-bacterial elements of the organism would come into oplication upon them. In the case of bacteria cultivating themselves the tissues, only those antibacterial elements would come into appli-

[1] *Lancet*, December 23, 1899. (pp. 36–44 *supra*).
[2] *Scientific Memoirs by Officers of the Medical and Sanitary Department of the overnment of India*, vol. xii, pp. 96 *et seq.*
[3] *Proc. Roy. Soc.*, vol. lxxiv, 1904, pp. 151 *et seq.* (pp. 103–105 *supra*).
[4] *Proc. Roy. Soc.*, B, vol. lxxvii, 1906 (pp. 161–164 *supra*).

cation which had passed out from the blood in the lymph in the regi
of infection. Further, inasmuch as lymph, coming in contact in successi
with a number of bacteria, or, as the case may be, with their product
would part with its antibacterial elements to those first encountere
retaining after percolating through a first bacterial nidus to a secon
or through the outer portion of such a nidus to its interior, only a residuu
of its original antibacterial power; there would come into existence,
particular in the case when the lymph-flow stagnated in the tissue
conditions far more congenial to the cultivation of bacteria than tho
which obtain in the blood.

Premising that I shall, as I proceed, ever and anon have to recur
the general principles enunciated in this first section of my paper, I ma
pass on now to consider the application of therapeutic inoculations
tubercle vaccine in connexion with the treatment of tuberculosis.
will be convenient to take up first the consideration of the treatment
strictly localized tubercular infections. Examples of such strictly localize
tubercular infections are furnished by most cases of lupus, further b
the large majority of tubercular invasions of the subcutaneous tissu
lymphatic glands, serous cavities, bone, testes, kidney, bladder, an
other internal organs, lastly by many apyrexial cases of phthisis.

PART II.

TREATMENT OF STRICTLY LOCALIZED TUBERCULAR INFECTION BY THE AID OF THERAPEUTIC INOCULATIONS OF A TUBERCL VACCINE.

In connexion with the treatment of cases of strictly localized tube
cular infection we have to take into account the following facts: (1
The tuberculo-opsonic power of the blood in these cases appears to b
uniformly inferior to that of the normal blood. (2) The immunisin
stimuli which are required for raising the opsonic power and for mair
taining it at a high level here make default. (3) The tubercl
bacilli are cultivating themselves in the focus of infection under cor
ditions which are much more favourable to their growth than those whic
obtain in the case of the circulating blood. (4) An increase of the opsoni
power of the blood can be achieved and maintained by the inoculatio
of a series of appropriately adjusted and interspaced doses of tubercl
vaccine. (5) We have at disposal methods by which we may increas
the lymph-flow through the focus or foci of infection in such a manner a
to bring the antibacterial elements of the blood into application upo
the invading bacteria.

It would be impossible within the limits of space within which I hav
here to confine myself to bring before you the evidence in support of a
the above propositions. So far as it relates to the first four of the abov
propositions, that evidence has been set forth in detail in a communicatio

the Royal Society made in conjunction with Douglas [1] and in a further nmunication made to the same Society in conjunction with Staff-rgeon S. T. Reid, R.N.[2] I may, therefore, here limit myself to the con-eration of the proposition that we have at our disposal methods for ding through the focus of infection a stream of antibacterial lymph.

cussion of the Means which are available for sending a Stream of Antibacterial Lymph through the Focus of Infection.

The activation of the lymph stream in the focus of infection supplies *rationale* of a number of procedures which have been empirically ctised. Our grandmothers were wont to activate the lymph stream boils—they spoke of it as "drawing the boil"—by the application of t poultices followed by sugar-and-soap plasters.[3] The surgeon of these ter days practises the method when he applies hot boric fomentations connexion with the treatment of septic infection associated with nphangitis. Professor Bier in Germany practises the method when obstructs the circulation in a limb or, as the case may be, in the head d neck, with a view to achieving an effusion of lymph. The physician, ake it, practises it in connexion with the application of his rubefacients d preparations of iodine, and possibly also when he administers expec-rants—deeming that he is only "loosening the expectoration." The ray, the radium, and the radiant heat therapists practise it in con-xion with the exercise of their particular professions. Lastly, as I ink emerges very clearly from the facts which Dr. Bulloch proposes to y before you, the work of the Finsen light therapist resolves itself o an application of this method.

It seems to me all that is further needed in connexion with these ethods is that they shall be employed purposefully as means to an end d not blindly as empirical methods. For I conceive that if this were ne it would immediately be recognized (*a*) that the douching of a bac-rial nidus with a rapidly flowing stream of lymph might in the case here that lymph possesses only very inferior antibacterial properties associated with risk; (*b*) that the irrigation would always be more fective in the case where the antibacterial power of the blood had pre-ously been raised either by auto-inoculation or artificial inoculation;) that an ampler lymph-stream could in every case be obtained by lministering decalcifying agents (such as citric acid) in such quantities might suffice to reduce the coagulability and at the same time the scidity of the blood; (*d*) that in the case where the focus of infection is ositioned in a lymphatic gland the blood stream might with advantage

[1] Wright and Douglas (Loc. cit.).
[2] Wright and Reid (Loc. cit.).
[3] Let us reflect in this connexion that the hot poultices determined the blood ream to the focus of infection, that the sugar by its osmotic power drew the lymph rough the open boil, and that the soap by decalcifying the lymph prevented it agulating and forming a scab and so blocking the outlet.

be determined not only to the lymphatic gland involved but also to whole territory which sends its lymph to that gland; (*e*) that in ca where the focus of infection is positioned in the skin, and where the blo supply to the skin is inefficient, advantage might be taken of any medici agent, such as thyroid extract, which increases the cutaneous blo supply; (*f*) that inoculation of old tuberculin may possibly find useful incidental application in certain cases of superficial lupus producing an outflow of lymph through the affected skin; (*g*) th the injection into discharging sinuses of a decalcifying agent dissolv in a concentrated salt or sugar solution may possibly be found useful causing an irrigation of such sinuses by lymph; and, lastly, (*h*) that the therapeutic effect of the Finsen rays should resolve itself merely in a question of determining lymph to the site of infection, it would be w in every case to preface it by inoculation procedures or, perhaps, even use in association with these last some cheaper and less laborious ther peutic device. I would throw out merely as a suggestion that we ha in the application of bags filled with hot sterilized sand [1] a very ine pensive and convenient method of determining a blood stream to a region on the surface of the body.

Digression on the Results of Ordinary Surgical Methods as applied t the Treatment of strictly Localized Tuberculosis.

Before saying what I have to say on the subject of the results obtain by treating strictly localized tubercular infections by the aid of therapeut inoculations of a tubercle vaccine I will, if I may, say a preliminary wo on the results as obtained by methods which are believed to be pure surgical, leaving the discussion of the results obtained by the Finse light treatment to be dealt with later in this discussion by Dr. Bulloch

It is the belief of the surgeon—one, I take it, of his cherished belie —that it is possible to extirpate completely and effectively by the knif in a large proportion of the cases which he undertakes, the invadin micro-organisms. I, for my part, find it very difficult indeed to believ that this result can often be achieved even by the most radical operation While I have great difficulty in believing that these means can be a effectual as they are claimed to be, I do not—though this may first fa upon your ears as a paradox—find any difficulty in believing that th good results which the surgeon claims are often achieved. But succes is, as I gather, obtained in some cases where the operation leaves some thing to be desired from the point of view of completeness; and again, a other times, the efforts of the surgeon come to nought, in spite of th fact that the operation has been conducted with scrupulous care.

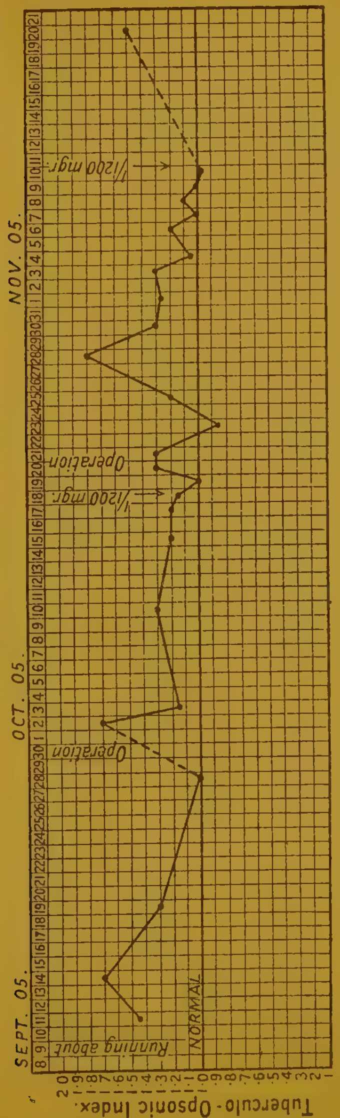
[1] For the sterilization of the sand I am accustomed to give the following instruc tions:

Place the sand in a saucepan over the fire, having previously stirred in a numbe of small pieces of white paper. Continue the stirring until, with the attainmen of a temperature of 200° C., the pieces of paper have all turned brown.

[2] *Vide also pp.* 142–149 *supra.*

Even if we leave altogether out of account the possibility that the tients successfully operated upon may have been patients whose tuber-lo-opsonic indices were previously to inoculation on the average higher

CHART 4 (by Dr. J. Freeman).



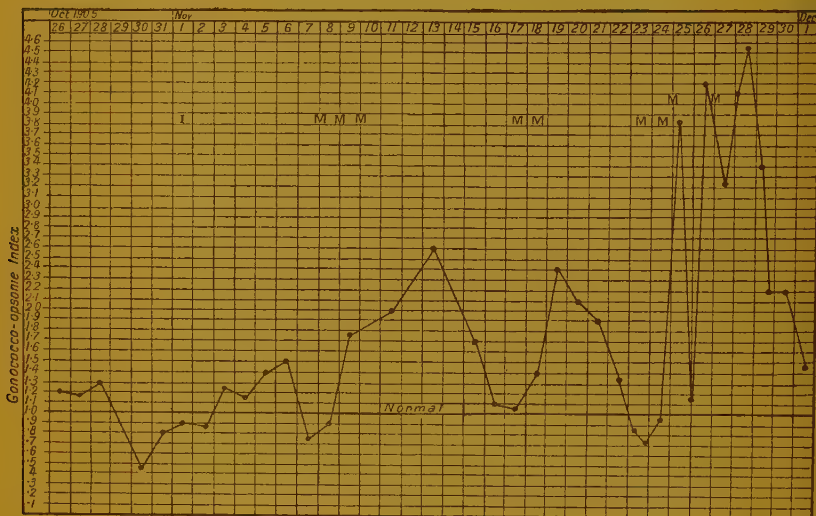
Shows, in the case of a child suffering from tubercular caries of the fibula, that the tuberculo-opsonic power may be raised by active exercise and surgical procedures as well as by the incorporation of tubercular elements in the form of therapeutic inoculations of new tuberculin.

an those of the patients unsuccessfully operated upon, the observations f my fellow-worker, Dr. J. Freeman, clearly show that there are other actors which may influence the result. There is, in the first place, the

possibility that surgical interference with a tubercular focus may, as i
the two scraping operations [1] which are in question in Chart 4 b
followed by the whole train of events which we have learned to associa
with the inoculation of a bacterial vaccine. Again, as in the initial rise o
the tuberculo-opsonic power which is set forth in the same curve, physic
exercise may be an active agent in connexion with the production o
immunity. Finally, as shown in Chart 5, massage of the focus of infectio
may produce effects similar to those of inoculation.

When we come to reflect upon the matter there is nothing in any o
this to surprise us. A conveyance of bacterial elements into the bloo

CHART 5 (by Dr. J. Freeman).



Shows the effect of massage upon the gonococco-opsonic power in the case of a man suffering from gonococcal arthritis. I, inoculation of gonococcus vaccine (50,000,000 gonococci). M, massage.

is precisely what might be expected from surgical procedures which open
up the lymph spaces, or as the case may be from the activation of the
lymph stream by kneading operations or ordinary muscular movements.
In view (*a*) of the observations of Meakin and Wheeler (to be referred to
below, p. 296) on the effect of physical exercise upon the tuberculo-opsonic
index in phthisis; (*b*) of the observations of Freeman, made in my labora-
tory, on the effect of massage on the opsonic power in the case of a variety
of bacterial infections; and (*c*) of the observations of Clarence Wright

[1] The same sequence of events has already been met with in connexion with three other cases. One here referred to was a scraping operation undertaken upon tubercular glands, the other two were extirpating operations also undertaken in connexion with tubercular glands. It would seem probable that similar results would be obtained also in connexion with curetting operations undertaken upon the uterus in connexion with bacterial infections.

rchives of the Röntgen Ray," December, 1905) on the effect of X-ray tment on the tuberculo-opsonic power in the case of lupus patients, iggests itself that it may prove possible to determine the nature of any lized bacterial infection by measuring the opsonic power of the blood h regard to the suspected micro-organism before and after massage, sical exercise, the application of X rays, or any other method which ivates the local lymph stream.

the Results which have been obtained by the Treatment of Localized Tubercular Infections by the Aid of Therapeutic Inoculations of a Tubercle Vaccine (Koch's New Tuberculin) controlled by Determinations of the Opsonic Index.

Returning from the above digression to take up again the main theme this discourse, and coming to the question of the results which ve been achieved by the therapeutic inoculation of Koch's new tuberin, safeguarded by systematic determinations of the opsonic index, nd myself face to face with the impossibility of conveying to you an equate idea of the results which it is possible to achieve in the matter the cure of localized tuberculosis. To convey to you such an adequate a I should have to bring you face to face with the patients and to reproce for you, in the case of those who were suffering from external lesions, eir past by the aid of photographic records. In view of my having been evented by external circumstances from securing photographs of the ses when they first presented themselves, and in view of the circumnce that the presence of the patients to-night would have broken in an unwarrantable manner upon this discussion, I must do what I can the bald method of narrative, inviting you hereafter to inspect the tients either at St. Mary's Hospital, or, if it shall be signified that this the wish of the Society,[1] here in this room when we resume this scussion.

Before entering into a recital of the cases, I would point out that in ch case a measurement of the tuberculo-opsonic power of the blood s (with only rare and isolated exceptions) been made in connexion th each inoculation of tubercle vaccine. The burden of the very many ousands of hours of work which this has involved has been shared with e in the most devoted and self-sacrificing manner by my friends and low-workers, Captain Stewart R. Douglas, I.M.S., and Dr. J. Freeman. vould therefore have it borne in mind that the work which I shall here mmarize is in very large part their work.

For the purpose of the summary I may classify the cases of localized berculosis, which we have dealt with under the headings of *lupus*, *bercular ulceration of the subcutaneous tissues and bone*, *tubercular glands*,

[1] In conformity with the wish of the Society a number of the cases whose hisries are chronicled below were exhibited at the meeting of the Society held on ecember 12, 1905. The cases thus exhibited are in the records below distinguished an asterisk (*).

tubercular disease of the genito-urinary system, tubercular disease of join *tubercular peritonitis,* and *apyrexial phthisis.*

While we have treated, or have under treatment at present at S Mary's Hospital, examples of each and all of these classes of cases, v have, keeping in view the importance of testing the method of inocul tion in the most rigorous manner, devoted our attention in particular cases where definite objective evidence could be obtained of any alteratic in the clinical condition, and by preference to cases where ordinary surg cal methods had already been unsuccessfully exploited. In conformit with this I shall, in the summary below, consider in particular the resul obtained in the treatment of the four first-mentioned categories of tube cular infection, and will pass over in silence the less convincing thoug equally satisfactory results which have been achieved by inoculation i the cases of tubercular peritonitis and joint disease which we have ha under treatment. I shall omit from consideration also the satisfactor results achieved in three out of the five cases of phthisis which we hav treated by tuberculin inoculation. It is to be borne in mind in connexio with this very small number of cases of phthisis treated that we hav except under very special circumstances, excluded this affection fro treatment, because of the difficulty of eliminating in an Out-Patien Department that class of phthisical patients who, being already th subject of auto-inoculations, cannot, it seems to be, except under ver special precautions, safely be treated by the method of inoculation.

Lupus.

If we except one of our very earliest cases—where the results of a fe weeks' treatment were, probably owing to the administration of to large doses of the vaccine, such as to discourage the patient,[1] and t lead him to abandon the treatment—we may say that the inoculatio treatment has, in all the cases of lupus we have dealt with, ameliorated but so far only in one case [2] cured, the disease. Not infrequently w have seen certain of the patches completely cured, while the disease i other regions has remained refractory. These only partially successfu results, which contrast in a very unfavourable manner with those obtained in connexion with tubercular ulceration affecting the deeper tissues, depend it seems to me, not so much upon a defective power of response to inocu lations on the part of the lupus patient as upon the inadequate manne in which the antibacterial substances come into application upo the tubercle bacilli in the case where these are disposed in a skin which is but poorly supplied with blood. I do not doubt that i it were possible to superadd to the treatment by inoculation anothe form of treatment which achieves, as the Finsen light appears to do, a sufficient transudation of lymph into the skin, the efficiency of the

[1] *Vide* Graham Little, *British Journal of Dermatology*, September, 1904.
[2] This case was shown to the Society on December 12, 1905.

culation treatment as applied to lupus would be much greater than has been in our hands.

Tubercular Ulceration of the Subcutaneous Tissues.

The clearest and most unfallacious evidence of the advantage which be derived from the therapeutic inoculation of tubercle vaccine can be nished in connexion with tubercular ulceration of the subcutaneous ues. It is not a question here of the achievement of success in a tain percentage of cases where ordinary surgical methods have failed. to the present, at any rate, it has been a question of *uniform* success. e following series of cases, all of which, except the last, are available inspection, furnish evidence of what can be achieved by inoculation cases which had defied all ordinary methods of treatment, and which ght quite well have been reckoned desperate.

CASE 1.*[1]—The patient, whose case I have already reported upon,[2] man of about thirty years of age. His history is as follows: In the umn of 1902 he developed tubercular glands on the left side the neck and a tubercular abscess on the point of the left ulder of the same side. He was admitted to St. Mary's Hospital and s operated upon for the first time in January, 1903. The wound oming invaded with tubercle and refusing to heal, further operative cedures were embarked upon. In all six successive scraping, extirzing, and skin-grafting operations were undertaken during the course the year, the wound becoming in each case reinfected, and the area of eration being extended. In December, 1903, when the patient came for treatment by inoculation, the whole area from the point of the shoulder to the base of the ear formed a single deep eroded ulcer. e lobule of the ear was half eaten away, and immediately underneath a deep ulcerated crater had developed, which looked as if it was going break into the pharynx. The adjacent side of the face was distorted swelling, giving the patient the appearance which would go along h a one-sided attack of mumps. The axilla was occupied by a gland ich was as large as a pigeon's egg, and the patient was haggard and y emaciated. After eight months' inoculation with new tuberculin, plemented on several occasions by inoculations of a staphylococcus ccine, and the local application of formalin gelatine,[3] I was able to ort that steady improvement had been made under the treatment, t the swelling of the face had almost entirely disappeared, that the ter under the angle of the jaw had healed up from the bottom, that e gland in the axilla could no longer be felt, that the ulcerated wound d almost entirely closed over, and that the patient might now almost

[1] The patients who are distinguished with an asterisk (*) were exhibited to the iety on December 12, 1905.
[2] *Proceedings of the Royal Society*, vol. lxxiv, July, 1904, and *Clinical Journal*, vember 9, 1904 (*supra*, p. 126, and pp. 265–266).
[3] *Vide* author's paper, *Lancet*, July 9, 1904.

pass muster as a healthy man. After a further three months I report that the wound was entirely closed and that there remained only area of the size of a threepenny-piece, which was still covered scab. I also pointed out that the previous site of the ulcer was cover in, not by scar tissue, but by a quite soft and elastic skin. Carryi on the history of the patient for another year, I have to report th after having got completely well, and after treatment had been di continued for about six months, he presented himself again for trea ment at the hospital. He had now, after exhausting work as a barma developed a very large soft gland in the previously sound side of t neck, and another in the groin of the same side. His opsonic ind was found to be very low. The glands in question rapidly broke dow leaving crateriform openings, which presented all the typical clinic appearances of syphilitic gummata. No improvement having manifest itself under a very thorough antisyphilitic treatment carried out in t hospital, and the patient's tuberculo-opsonic index ranging always abo 0·4, the tuberculin inoculations were recommenced, with the result th he is now making a marvellously rapid recovery.[1]

Case 2.*—The patient, a woman, aged thirty-one, has, like the las been previously reported on.[2] Her history is as follows: She developed tubercular infection of the glands of the neck at the age of fourteen year Then suppuration supervened and the abscesses were opened, the wounc became infected, and other glands also became involved. Later, tube cular disease developed in the little finger of the right hand. The tw terminal joints of that finger were removed fifteen years ago, when th patient was sixteen years old. About this time lupus broke out c her face and on her left arm and hand. At the age of nineteen years sh underwent treatment with Koch's original tuberculin. She received fro three to four inoculations a day (the total of her inoculations amountin to 150). This treatment resulted in violent inflammatory reaction i the patches of lupus, a piece of bone sloughed out of her left arm, and sh remained in hospital seriously ill for thirteen weeks. She attributes, an no doubt rightly, the aggravation of symptoms and ultimate loss of he arm to these inoculations. We can now discern that there must hav been produced a cumulative negative phase. After a respite vigorou treatment was resumed in another hospital. The lupus patches wer then frequently scraped and many glands were extirpated from the necl In 1900 the Finsen light treatment was resorted to and was persever in for eighteen months. This effected superficial improvement in th condition of the face and neck, but the disease continued to spread in th deeper structures and in particular in the bones of the left arm. Finally it became necessary to amputate this limb. The disease now re-invade the stump and broke out in the point of the shoulder and in the fron

[1] *This patient is now perfectly well and has since* (1907) *been actively engaged a one of our laboratory assistants.*

[2] Loc. cit. (*supra*, pp. 127–128, and pp. 266–267).

the chest. Röntgen rays were now tried unavailingly. Finally, in cember, 1903, the patient, who was then in a very reduced physical dition, was referred to me by Dr. E. G. Graham Little for treatment by erculin inoculations. I was able to report in November, 1904, that patient had arrived at a tolerably satisfactory condition in the matter her general health. Her body weight had gone up and had reached ½ lb., as much as 5 lb. having on one occasion been gained in the interval ween two successive inoculations. The discharge from the sinus r the sternum and from the sinus in the stump of the left arm had ctically ceased. I am to-day able to state that the patient is in ust health, that the discharge from the sinuses has entirely ceased, and t she has been able for months to make use of an artificial limb. cept for some superficial patches of lupus on the face, she may be said be perfectly well.

CASE 3.*—The patient, a female, aged twenty, has also been previously orted on.[1] When she presented herself for treatment in December, 03, she had the appearance of a child. Her bones protruded ough the skin of her back somewhat after the fashion in which nes protrude in dried fish. The point of her nose was covered h a thick mass of scabs superposed upon a very angry-looking ch of lupus. The angle of the jaw and the front of the neck re occupied by patches of lupus in a similar condition. Both her t and her hands were affected with lupus. Her hands in particular re a mere mass of ulceration, the bones of the hand being also affected many places. The patient has made slow but steady progress under the culation treatment. The ulcers on her right hand are nearly healed, l those on her left hand considerably amended, and her general physique s improved. The patches of lupus on the front of her neck and under angle of the jaw are now represented by perfectly sound cicatrices, l the patch of lupus on the nose is improving.

CASE 4.*—The history of this patient, who has been under the care Mr. H. Stansfield Collier, is as follows: In 1900, at the age of thirty, right testicle and a gland in the groin were removed on account of ercular disease. Early in 1903 an abscess was opened some distance ove the ankle on the outer side of the right leg. The wound did not l. In December another abscess had formed over the external mallus, and a considerable portion of the lower end of the fibula was gouged ay. In July, 1904, another abscess developed in the lower third of leg and was opened. Just before the patient was taken over for atment by inoculation in January, 1905, the amputation of the foot s regarded as almost inevitable. The condition was as follows: A us wide enough to take a large drainage-tube led through the leg behind ankle-joint. A deep ulcerated trench occupied the region of the irpated fibula, and extended under the inner malleolus for a considere distance. A gland the size of a bantam's egg occupied the right

[1] P. 128 and 267.

groin. Rapid improvement set in almost immediately after the inaugura-tion of the inoculation treatment, the wound healing rapidly and th gland in the groin disappearing. The patient left hospital on the hig road to recovery toward the end of June. With the continuation the treatment the ulcer entirely healed, the whole affected area bein occupied, as it is now, by very soft elastic skin which does not in any wa interfere with the movements of the foot. Towards the end of Septembe 1905, a small swelling developed in connexion with what had been th upper border of the ulcer. This was opened and scraped, and the patien is now practically well, except for the fact that the scar of the last incisio is still covered with a very delicate scab.

CASE 5.*—The patient, a woman, aged twenty-eight, presented herse for treatment in October, 1904, with tubercular ulcers on her legs, whic dated back to her fourteenth year. These had been treated by antiseptic of various kinds for thirteen years, and had been scraped and skin-grafted On the right leg the ulcerated surface corresponded in its dimension with a five shilling piece. Around this was an extensive area of thi glossy skin. The ulcer on the left side occupied an area which extende from a little below the level of the ankle to nearly the middle of the leg In this area the tendo-Achillis and the peronei tendons were laid bare and the point of the foot was drawn down so that the toes alone came i contact with the ground. All round the ulcerated area the skin of th leg was thin and glossy. The patient's tuberculo-opsonic index worke out as 0·17. After admission to hospital the patient was treated wit therapeutic inoculations of tuberculin, supplemented by occasiona staphylococcus inoculations and the local application of formalin gelatine After six months' treatment the ulcer on the right leg had completel healed, and that on the left leg had been reduced to comparatively smal dimensions. The inoculation treatment, which had raised the tuberculo opsonic index of the blood to 1·8, and which had maintained it generall well above 1, was now supplemented by skin-grafting, and the patien left hospital with the ulcers completely healed, and looking a picture o health. This continued until some six weeks ago, when, in associatio with a sinking away of the tuberculo-opsonic index to 0·8, a small vesicle developed on the inner side of the leg at the border of the healed ulcer This broke down into a superficial ulcer corresponding in dimensions to a lentil. In association with an improvement in the tuberculo-opsonic power obtained by minute attention to dosage and proper interspacing of the inoculations, extension of this small ulcer has been arrested.

CASE 6.*—The patient is a man aged thirty-five. When he presented himself for treatment in July, 1904, he had been for two years the subject of an inflammatory knobby tumefaction of the subcutaneous tissue in the region of the jaw and over a considerable area of the throat. The case had been diagnosed as actinomycosis and had been treated without result by scraping and iodide of potassium. The patient's opsonic index with respect to the tubercle bacillus was 0·67. With respect to the staphy-

cus it was 1. The patient is now, after fifteen months' inoculation very small doses of tuberculin, nearly well. Throughout the course treatment it has been brought out very clearly (*a*) that the clinical tion corresponds in a very accurate manner with the tuberculo-ic index, and (*b*) that the patient's opsonic index can be maintained much higher level when doses in the neighbourhood of $\frac{1}{80000}$ c.c. of are inoculated than when larger doses are employed.

ASE 7.*—The patient is a man, aged about thirty-five, a furrier. he presented himself last June for treatment the dorsum of his was occupied by a deep ulcer corresponding in dimensions with a ized watch and surrounded by a raised edge. The ulcer had been ed by scraping. The patient's tuberculo-opsonic index stood at 0·85. d improvement both in the opsonic index and in the clinical con-n followed upon the inoculation of tuberculin, supplemented, when appeared desirable, by the inoculation of a staphylococcus vaccine. ulcer has now entirely healed, and the site of the ulcer is covered in soft and elastic skin, which does not in any way impede the move-s of the fingers.

Tubercular Invasion of the Lymphatic Glands.

ext, perhaps, to tubercular ulceration of the subcutaneous tissue, rcular affections of the lymphatic glands furnish the clearest evidence e efficacy of therapeutic inoculation of tubercle vaccine. This result, flection will show, is in accordance with what might have been ted *à priori* in view of the fact that the tubercle bacilli are here sed right in the path of the lymph stream, which is passing back gh the gland to the blood. I do not myself doubt from what I seen of the effect of inoculation on tuberculous glands that the pation of these by surgical methods, as well as the purely climatic ment of this affection, are destined to give place to the therapeutic itation of tuberculin inoculations, controlled by the determination he opsonic index, and combined with hot sand poultices and facients, or other measures which, like these, will produce an er lymph flow in the whole territory—or may I call it "watershed" collecting basin"—whose lymph passes into the blood through onduit of the infected gland.

ASE 1.—The patient was a young married woman who had undergone e hands of two distinguished London surgeons three successive ations for the extirpation of glands. When she presented herself reatment in January, 1904, three or four glands could be felt in the , the largest one being of about the size of a small walnut. After iagnosis of tubercle had been confirmed by a test inoculation, under-n with Koch's old tuberculin, the vaccinal treatment with the T.R. rculin was inaugurated. Three months later, after six inoculations, ucted with doses which were gradually increased from $\frac{1}{50000}$ c.c.

to a maximum of $\frac{1}{5000}$ c.c. the glands could no longer be felt a the dragging pains in the neck had entirely disappeared. After the dis pearance of the glands a few more reinforcing inoculations were giv In January, 1905, the patient presented herself again, with swelling the glands that had been previously affected. This swelling was, before, associated with dragging pains, and the patient was thinking having recourse again to operative procedure. In lieu of this the tub culin inoculations were resumed, with the result that after three ino lations, undertaken in the course of a month, the glands again co pletely disappeared. They have, so far as I can learn, given further trouble.

Case 2.—The patient was the wife of a medical man. She h suffered from childhood from swollen glands on one side of the ne These had become the source of constant dragging pains, and the larg gland, situated under the angle of the jaw, was large enough to prod considerable disfigurement. After three months of the inoculation tre ment the glands had much diminished in size, and no longer gave rise any disfigurement or discomfort.

Case 3.—The patient, who had been a nurse and who had alrea undergone two operations at the hands of a distinguished surgeon, v referred by him for treatment in April, 1904, with a recurrence of tub cular glands in the neck. After three months' inoculations the swell in the glands had entirely subsided. In association with this there v a very marked improvement in the general health. By the desire of patient the inoculations are still being continued as a precaution agai further recurrence.

Case 4.—The patient, a girl, aged four, came under treatment connexion with a recurrence of glands very shortly after operatio Her tuberculo-opsonic index stood at 0·7. After a series of six or sev inoculations the swelling in the glands had entirely disappeared.

Case 5.—The patient, aged four, came under treatment at the e of June, 1905, in connexion with a recurrence of tubercular glands af operation and continued discharge from a deep gaping wound in t submaxillary region. By the end of September, after a course of tub culin inoculations, supplemented on one or two occasions by an inoculati of staphylococcus vaccine, the wound had completely healed over a the glands were notably diminished in size. By the end of October tre ment was discontinued, the swelling in the region of the wound and the glands having entirely disappeared, the child being in absolute robust health.

Tubercular Disease of the Genito-Urinary System.

From some points of view more convincing, in others only less co vincing, than the results obtained in connexion with lesions which a

rectly accessible to sight and touch, are the results obtained in connexion ith tubercular disease of the genito-urinary system, in particular in the ıses where this involves the bladder. We have, in the fact that these ıses are associated with distressing pain and frequency of micturition, ıd in the fact that the presence or absence of tubercle bacilli in the rine can here be determined by microscopic observation, the means of easuring success and failure.

CASE 1.—The patient, a man aged twenty, when first seen twelve onths ago was suffering from extreme frequency, and looked worn with ain. He was only with difficulty able to draw himself upright, and could ıly with some distress climb upstairs. There were considerable swelling ıd tenderness in the prostate and back of the bladder, and the urine ontained some blood and a large quantity of pus. Microscopical examiations revealed tubercle bacilli in considerable numbers in the urine. ultures showed that there was no other bacterial invasion. The atient had been previously treated with inoculations of T R., the oses having been increased by geometrical progression up to $\frac{1}{2500}$ c.c. fter the inoculation of the larger doses the pain and frequency of icturition were greatly aggravated. After waiting till the immediate fects of the last inoculation had passed off, inoculation was recomenced with $\frac{1}{10000}$ c.c. of T.R. The tuberculo-opsonic index of the lood now stood at 0·62. After repeating the inoculation within $\frac{1}{10000}$ c.c. intervals of ten days, and then tentatively advancing to a dose of $\frac{1}{000}$ c.c. without achieving any sensible improvement in the opsonic ıdex or clinical symptoms, the dose was reduced to $\frac{1}{80000}$ c.c. The oculation of this dose at ten-day intervals was followed by steady ıd sustained improvement, both in the opsonic power and clinical mptoms. There was also a marked diminution in the prostatic tumection. After the dose had been for a time increased to $\frac{1}{60000}$ c.c. was again reduced to $\frac{1}{70000}$ c.c. While the frequency of micturion and the prostatic swelling have been much abated, and while the atient is practically free from pain, his urine still contains tubercle acilli. His condition is, however, now such that he is able to hold is urine for two hours at a time, and capable of undertaking without tigue a long day's shooting.

CASE 2.—The patient is a young woman of very good physique. She ame under treatment first in January, 1905, with a history of tubercular ystitis and tubercular disease of the kidney dating back two years. ne of her kidneys had been removed and there was evidence of the ıvolvement of the other kidney. The urine contained pus in considerble amount, and in association with this many tubercle bacilli and several arieties of contaminating bacteria, among others *proteus*, The patient's ıberculo-opsonic index was tested on two occasions before the inoculation eatment was initiated. On the first occasion it stood at 0·75, on the econd occasion at 0·35. An improvement in the patient's symptoms et in practically immediately after the first inoculation, undertaken with

$\frac{1}{80000}$ c.c. The tuberculo-opsonic power rose on the day after oculation to 1·7, and continued at this height or near this point for next six days. An inoculation undertaken on this day with $\frac{1}{40000}$ brought down the opsonic power of the blood. In association w this the patient complained of more pain. After the inoculations been continued for about six months, when the tubercle bacilli had dis peared from the urine, and when, as a result of the inoculation of a prot saccine, the *proteus* also had disappeared, the patient felt so well t hhe mooted the question of engagement and marriage. Since then vas suffered a relapse, developing an acute cystitis. This attack, wh was apparently associated with a reappearance of the *proteus* in the uri is now subsiding.

CASE 3.—The patient, like the last, is a young woman of good physiq who, after suffering from pleural effusion and severe cramps in the loi suddenly in December, 1904, developed severe cystitis and haematur Tubercle bacilli were now found in the urine. When she came un observation in September last she was suffering from great frequency micturition (up to twenty-five times in the night) and pain. Tuber bacilli were sparingly present in the urine, while a form of pneumococ was abundant. Her tuberculo-opsonic index stood on the first exami tion at 0·85 and on two subsequent occasions at 0·9 and 0·93 resp tively. Inoculation was begun with a dose of $\frac{1}{80000}$ c.c. of T.R. b this dose appears to have been excessive, inasmuch as the patient spe after inoculation a week of misery, the frequency of micturition risi on one occasion to thirty-two times in the night. The dose e ployed in the next inoculation was $\frac{1}{240000}$ c.c. and this dose has be employed since with very satisfactory results, pain being grea diminished and frequency of micturition now averaging five times in t night, while an opsonic index of 1·6 has been achieved.

CASE 4.—This case has already been reported on by me twelve mont ago as follows [1]: The patient, a married woman, aged forty-three, was a mitted to hospital under the care of Dr. D. B. Lees in the middle of Marc 1903, complaining of frequency associated with severe local pain micturition and dragging pains in the loins, in particular on the l side. The urine contained pus, epithelial casts, and tubercle bacilli such numbers that they could be demonstrated in large clumps in eve field of a microscope in preparations prepared from the urinary sedime Examination of the bladder revealed the existence of a large ulcer. T kidneys were enlarged and tender ; the left one in particular was affect and suspicious signs were detected in the apex of one lung. Tubercu treatment was begun in the middle of April, 1903. The effect exert upon the body weight during the period the patient was in hospital exhibited in the figures below :

[1] *Clinical Journal*, November 9, 1904, p. 264 *supra*.

	Pounds.		Pounds
April 20	91	June 15 . . .	103½
„ 28	96	„ 22 . . .	105
May 11	92½	„ 29 . . .	107
„ 19	93	July 6 . . .	109¾
June 1	96	„ 13 . . .	107¼
„ 8	101		

The drop of body weight which is recorded on May 11 coincided on ıe one hand with the development of increased local pain and symptoms f giddiness and flushing, and, on the other hand, with a rapid fall in the gglutinating power of the blood, which is displayed in Curve 1 (*supra*, . 271). These were all, I take it, symptoms of the supervention of a umulative negative phase dependent upon a too hasty inoculation of rogressively increasing doses of vaccine. After leaving the hospital in uly, much alleviated, in the matter of pain and frequency of mic-urition, the patient attended as an out-patient, and under the treat-ıent her weight in September, 1903, reached 119 lb. The tuberculin ıoculations were continued up to July, 1904. All this while the tubercle acilli, which were examined for almost every ten days, became gradually ess numerous. By May they had completely disappeared from the rine. The patient none the less still suffered from serious bladder rouble—due, as appeared on examination, to cicatrization and great hickening of the bladder walls, and possibly to some superadded ulcera-ion referable to septic invasion by the *bacillus coli* and by a Gram-stain-ıg diplococcus—micro-organisms which have been throughout present ı millions in her urine. Taking up her history from this point, I may dd that shortly after the publication of the above report the patient ame back with symptoms of a relapse, and tubercle bacilli were once ıore found in her urine. Under the influence of further inoculations of ıberculin these again disappeared from the urine, and she continues to be ree from pain and is in very good health. She suffers, however, from ıcontinence.

Case 5.—The patient is a man of some forty-five years. His history is as ollows: In 1904 the right testicle became swollen and an abscess formed hich left behind a sinus in the posterior aspect of the scrotum. In July, 905, when the patient came into hospital, the right testicle was found to e typically tuberculous, and there was discovered also a small nodule ı the left epididymis. The patient was unable to hold his urine for more han half an hour at a time night and day, and in association with this ubercle bacilli were found in the urine. He was now treated by inocula-ions of tuberculin and left hospital in September much improved. Since eaving hospital he has put on 14 lb. in weight, and the frequency of ıicturition has been reduced, averaging now only four times in the night.

ummary of the Results obtained by Therapeutic Inoculation in Cases of Localised Tuberculosis.

In view of the very favourable and, what is almost more important,

uniformly successful results which can, as will have appeared, be obtain even in the most intractable cases of localized tubercular infection the therapeutic inoculation of tuberculin carried out under t safeguards explained above, and in view of the fact that not l favourable results can be obtained by the aid of the correspondi bacterial vaccines in the treatment of localized infections by oth micro-organisms, I do not hesitate to contend that we have, in the pow of raising the anti-bacterial power of the blood with respect to any i vading microbe, out of all comparison the most valuable asset in medicine

I would, in view of this new asset in medicine, fain induce t surgeon to abate something from his conviction that extirpation a the application of antiseptics offer in connexion with bacterial infection t only possible means of cure ; I would have the surgeon resort to extirp tion only when the physician tells him that all other means have be exhausted ; and I would have the physician assume everywhere t rôle of an immunisator ; and I would have him defer handing over h patients to the surgeon before he has tried in every case of localized bacteri infection which is unassociated with immediate risk to life the therapeut inoculation of the appropriate bacterial vaccine.

PART III

TREATMENT OF SYSTEMIC TUBERCULAR INFECTIONS BY THE THERAPEUTIC INOCULATION OF A TUBERCLE VACCINE

In connexion with pyrexial phthisis and other forms of pyrexi tuberculosis we are face to face with a problem which confronts us als in connexion with every other systemic infection—i.e., the problem a to whether in view of the fact that the machinery of immunisation already spontaneously called into action, any advantage can be looke for from the inoculation of bacterial vaccines. Before an answer can b given to the question, we must try to form to ourselves (*a*) some con ceptions of the conditions with which we have to deal in ordinary systemi infections such as are represented by continued fevers, and again (*b* some conception of the special conditions which we have to deal with i pyrexial phthisis and similar tubercular affections.

Conditions which we have to take into Account in connexion with Ordinary Continued Fevers.

In the case of continued fevers, such as typhoid fever and Malta feve in man and anthrax in animals, bacterial elements are passing more o less continuously into the blood from regions like the spleen, where th micro-organisms are cultivating themselves in close relation with th blood vessels. These bacterial elements, the nature of which we nee not pause to discuss, exert upon the organism not only toxic effects, bu call forth also immunising responses in all respects similar to those which are called forth by the inoculation of bacterial vaccines.

Where the influx of the bacterial elements which here come into nsideration does not exceed a certain maximum, there can be registered, doubt often after the intervention of a negative phase, a definite rease in the antibacterial substances in the blood. Such an increase registered, for instance, in connexion with typhoid fever and Malta er when, after the fever has persisted for some days, an agglutinative ction greater than that which is obtainable with the normal blood is tained. By the aid of the antibacterial elements (of which the agglu-ins furnish only the most easily demonstrated and therefore the most niliar examples) the invasion of the blood stream is checked. If now the continued elaboration of the antibacterial elements the bacterio-pic pressure of the blood is brought up to, and is sustained at, a suffi-ntly high level, the antibacterial effect makes itself felt, not only in e actual blood stream, but in the backwaters of the circulatory system, ere the blood flows comparatively slowly, and in the end also in e tissues. The invading micro-organisms will be finally disposed of en a lymph, rich in antibacterial elements, floods through all the foci lowered bacteriotropic pressure in which the bacteria are ensconced.

Alongside of the cases which run their course to this favourable con-usion there are other cases where the influx of bacterial elements into the ood is immoderate and uninterrupted. Here we may fear that, owing to a ralysis of the machinery of immunisation, the antibacterial power of the ood will not be sustained, and that the invading bacteria will establish emselves in the circulating blood. The possibility that, by the incor-ration of a bacterial vaccine into the body of a patient who is already aggering under a severe bacterial intoxication, such a further quantum of ison might be added as would just suffice to overtax his power of resist-ce is a risk which has to be considered in connexion with all therapeutic oculations of bacterial vaccines, undertaken in connexion with systemic fections. That risk may, according to circumstances, be very grave insignificantly small. Let me take the extreme cases. When, as in a se of fulminating typhoid fever, the system is profoundly intoxicated, and hen the absence of the agglutination reaction and the diminishing content the blood in other antibacterial substances show that the patient's ower of immunising response is probably already overtaxed, I presume at no one would like to take the responsibility of incorporating a further antum of bacterial poison. In the contrary case of a comparatively ght attack of Malta fever, where the fever is likely to run on for months ithout any serious intoxication of the system, and where the imperfect evelopment of the agglutination reaction seems to indicate that the nmunising impulses are making default, I have gladly taken upon myself, nd have counselled others to take upon themselves, the responsibility f applying further immunising stimuli in the form of a carefully safe-uarded series of inoculations. In each such case the event has justified e procedure.

The cases last considered have a direct application in connexion

with the question of undertaking inoculations of a tubercle vaccine connexion with pyrexial phthisis. I would point out in this connexi that there is, as between inoculations of bacterial made into the su cutaneous tissue and the introduction of bacterial poisons directly in the blood stream, a very important difference, which must be kept view. If the vaccines were to be incorporated directly into the blo stream, we should thereby contribute directly to the intoxication the central nervous system and the heart, and we should, as the ve disappointing results which are achieved in the immunisation of horses the intravenous injection of diphtheria toxins clearly show, be advanci but little in the direction of immunisation. On the contrary, when ba terial vaccines are incorporated into the subcutaneous tissues, and wh they therefore come into application in a concentrated form upon thes and are held fast by these, we may quite well be effecting a great de in the way of immunisation without contributing in any appreciab manner to the intoxication of the central nervous system and heart.

Special Conditions which we have to take into Account in connexi with Pyrexial Phthisis and other Localized Tubercular Infectio which are associated with Pyrexia.

In the case of localized tubercular infections which are associate with pyrexia, the conditions are different from those in the continue fevers which have been under discussion, (*a*) in the respect that the influ of the bacterial elements into the blood takes place from regions whic do not stand in immediate relation to the blood stream, and (*b*) in th further respect that this influx is discontinuous and stands in definit relation (to an extent which does not hold good in the case of other con tinued fevers), with causes which are to a quite appreciable degree unde control. The causes which come into consideration here are, in particula physical exertion, and mental effort, or excitement. Under the influenc of these causes there can be registered, not only a rise of temperatur but also a variation of the opsonic power similar to that which is encountere in connexion with the inoculation of tubercle vaccine. I may give a instances of such variations the cases of two phthisical patients, who too part in a dance, with the result that they both became ill, and tha their opsonic indices, which had never previously been found lowe than 1, declined to 0·12 and 0·33 respectively; further the case o another phthisical patient whose opsonic index fell in connexion wit overwork to 0·2 from a level of over 1. I may refer you also i this connexion to a paper [1] by my friends Dr. H. Meakin and Dr. C Wheeler, which records similar effects produced in the case of phthisica patients in connexion with walking.

We have a very simple explanation of these facts if we suppose that, under the influence of the limb and chest movements, and the circulatory

[1] *British Medical Journal*, November 25, 1905.

isturbance and increased lymph flow which are associated with excite-
ient, physical exercise, or mental overwork, tuberculous poisons from
ie infected tissues are conveyed into the blood. Especially may an
ıflux of lymph, loaded with tuberculous poison, be expected in the case
here the patient who is the subject of a tubercular infection of the leg
ndertakes walking. We have already in Chart 4, *supra*, p. 281, seen in con-
exion with a case of tuberculous disease of the leg a rise in the tuberculo-
psonic index which was to all appearances the result of running about.
have also quite recently seen another case of a similar association in
he case of a boy with tuberculous hip disease who, after lying on his
ack for years with a normal temperature, developed pyrexia on begin-
ing to walk. No doubt this boy conveyed into his blood as he walked
stream of lymph which had passed through his old tubercular focus.
n connexion with, and probably as a sequela to, this, I registered a
uberculo-opsonic index of 1·4.

'ossibility in connexion with Pyrexial Phthisis and other Localized Tubercular Infections which are associated with Pyrexia of quieting the Circulation and staunching the Lymph Stream in such a way as to arrest the Auto-Inoculations, converting the Systemic Infection in this manner into a purely Localized Infection.

The fact that in pyrexial phthisis and other localized tubercular
nfections which are associated with pyrexia the bacterial poisons are not
enerated in direct relation with the circulatory system is, as reflection
vill show, a fact which is pregnant with all-important consequences in
onnexion with the therapeutics of the systemic infections we have here
n view. While it may be possible in the cases of continued fevers like
yphoid fever to effect something in the direction of reducing the severity
of the intoxication and the dosage of the auto-inoculations by keeping
he patient perfectly quiet, the abolition of the intoxication and the arrest
of the auto-inoculations constitute in the case of phthisis not a remote
deal but an ideal which is every day realized. The complete rest in bed,
which gradually reduces the temperature in the large majority of cases
of tuberculous phthisis, as well as in other localized forms of tubercular
nfection, is, I take it, to be regarded as a therapeutic measure for making
n end to those auto-inoculations which follow upon every over-exertion,
nd which make the life of the phthisical patient, when abandoned to
is own devices, what my fellow-worker, Dr. R. H. Urwick, has shown
t to be,[1] to wit, a succession of negative and positive phases. I take
t that the rest in bed might, with a view to further staunching the lymph
low, with advantage be supplemented in every case by the administration
of therapeutic agents which will increase the coagulability and viscidity of
he blood. I have no doubt that this object is already in many cases

[1] *British Medical Journal*, July 22, 1905.

undesignedly and unwittingly attained by placing the patient on a dietary of milk.[1]

Consideration of the Question as to how far the Patient has been brought in the Direction of a Cure when his Pyrexia has been abolished, and his Auto-Inoculations have been arrested by Confinement to Bed.

We may usefully ask ourselves exactly how much will have been achieved in the case of a tubercular infection if, when the influx of tuberculous poison into the blood has been arrested and the pyrexia has been abolished, we stop at this point. The question is an all-important one in view of the years and years of complete inaction to which many patients are condemned on the theory that they are, while they continue to rest and wait, every day making progress in the direction of a cure. To any one who has surveyed the tuberculous patients laid out on spinal chairs in our seaside health resorts—waiting; or the patients who are lying in bed or upon deck chairs in our open-air sanatoria—waiting; it is plain as demonstration can make it that there is gained for the patient, by the arrest of the influx of tuberculous poison into his blood, a power of assimilating his food and an appearance of vigorous health. If only to the appreciation of this fact there could be added the belief that the cure of bacterial infections depends neither upon the storage of fat, nor upon the bronzing of the skin, nor yet upon the breathing of fresh air (sea-coast air, country air, pine-wood air, mountain air, or warm southern air), but only upon the destruction of the invading bacteria by the antibacterial substances of the blood (with or without the co-operation with the leucocytes), we should, I think, have come close to the truth.

It is, at any rate, my belief that with the cutting off of the auto-inoculations progress in the direction of immunisation is arrested, and that with that arrest the blood reverts in every case to the inferior level of antibacterial power at which the blood of the subject of strictly localized tubercular infection normally stands. If this is so—and I infer from what I have gathered that this is the condition of affairs in phthisis when it has by rest in bed been brought back into the condition of a strictly localized infection—it is eminently comprehensible that the patient is liable to relapse when on return to work he over-exerts himself in such a way as to convey into his blood—as happened in the case of the tuberculous disease of the hip before adverted to—tuberculous elements from foci in his tissues in which the tubercle bacillus has survived.

Programme of Treatment which would appear to be indicated in the case of Pyrexial Phthisis, or other Localized Tubercular Infection which may be associated with Pyrexia.

If the views which I have developed above are in harmony with the facts, the following programme would seem to be marked out for us in

[1] *Vide* author's paper on "Milk as a Medicinal Agent." *Lancet*, October 14, 1905.

onnexion with every case of pyrexial phthisis: (1) Our first efforts ught to be directed to bringing back the infection to the condition of a urely localized infection. Rest in bed and the adoption of measures or increasing the coagulability of the blood would be the appropriate ethods for the achievement of this end. (2) As soon as this first object as been achieved it should be our aim to substitute for the inappropriately djusted and inappropriately interspaced auto-inoculations which wore own the patient without achieving effective immunisation a system of ppropriately adjusted and appropriately interspaced inoculations of a ubercle vaccine. (3) Finally, as soon as by the means just indicated a atisfactory antibacterial pressure has been achieved in the blood, it hould be an object of endeavour, by the regulation of the patient's xercises and by attention to his blood pressure and by taking steps here necessary to diminish the coagulability of his blood, to irrigate in a ethodical manner all the foci of infection with a lymph rich in antibacterial substances.

A Criticism of the Foundations of Serum Therapy.[1]

Being a Contribution made to a Debate held under the Auspices of th Chelsea Clinical Society.

By A. E. Wright.

Preliminary Remarks—Incidental Disadvantages of Serum-therapy—Productio of the "Serum disease"—Treatment of the Serous Hæmorrhages (Urticari and Articular Pains) which are associated with the serum-disease by Adminis tration of Calcium Salts—Theoretical Basis upon which Serum-therapy i founded—Physician relies on the Bacteriologist who produces the Serum—H in his turn proceeds upon the unjustified assumption that the Animal which is vicariously inoculated will make unlimited response to Inoculations o Bacterial Vaccines—Real Course of Events after the Inoculation of Bacteria Vaccines—Possibility that the Sera as sent out may contain Bacterial Toxins— Question of responsibility for this—Question as to how presence of Toxins in Serum can be discovered—Laboratory experiments, Clinical results—Suggestion that a Therapeutic Serum may be a Vaccine in Disguise, and question as to whether the assumption that a Vaccine might be of service in Septicæmic Infections is a tenable hypothesis—Conclusion that this is a tenable hypothesis, and that it may explain the occasional Clinical benefit obtained in Septicæmic Infection by the Inoculation of certain Sera—Suggestion that Professor Chantemesse's successful Serum-therapy in Typhoid may be explained in this manner —Summary of Conclusions.

We are met this evening to discuss together the interesting question of serum therapeutics. Professor Hewlett has already, in his opening address, taken you over the ground, and you will have noted that he has said little in praise of the serum therapeutics as a general principle of treatment. Not only had he little to say in the way of commendation for serum-therapy as a method of combating bacterial disease other than diphtheria, but you will have noticed that much that he has said has been of the nature of extenuation and explanation of failure, coupled with suggestions for the modification of the various sera with a view to rendering them efficacious. I have as I listened felt myself in general agreement with Professor Hewlett on the question of what has been actually achieved by serum therapeutics On the other hand, I cannot feel that

[1] Reprinted from the *Clinical Journal*, May 16, 1906.

ofessor Hewlett's explanations of the failure of serum therapeutics connexion with the treatment of bacterial invasion go to the root of the atter. I have, for instance, no confidence whatever either in the acticability or in the utility of the suggestion that anthropoid apes, d generally speaking animals other than those now used, should be nployed to furnish the therapeutic sera, and I am not satisfied that ere is a basis of experimental fact for the theory which assumes that ra which are at present impotent would be rendered potent for good if ey were exhibited in combination with them—that "complement" hich is so often on the lips of bacteriologists.

I do not believe that serum therapeutics as applied to the destruction bacteria in the body can be bolstered up by any of the devices that ve been referred to. The whole system of serum-therapy, except here it is a question of the neutralization of a poison like diphtheria xin by the aid of an antitoxic serum, appears to me to rest upon very secure foundations.

Before attempting to make good this proposition I would like to take e opportunity of saying a word on the question of certain altogether cidental disadvantages which attach to serum-therapy in all its forms, en where, as in the case of the antitoxic serum which is employed in e treatment of diphtheria, the serum accomplishes all that it is designed accomplish.

You will immediately appreciate that I have in view those pathological enomena, such as urticaria and articular pains, which, since they ay occur after the incorporation of any foreign serum without dis-nction, are by the Germans very conveniently grouped together under e name of the *serum disease*.

The pathology of the "serum disease" is full of interest both from e theoretical and the practical standpoint. Viewed from the theoretical andpoint, the disease is interesting by reason of the fact that the henomena of the disease are, as is brought out clearly in the recent ork of Pirquet and Schick,[1] to be interpreted as events in a process of nmunisation by which the organism purges itself from the foreign rum. The serum disease is interesting also from the practical stand-oint, first because there is some reason to think that the process of urgation just referred to may be associated with fundamental alter-tions in the antibacterial power of the blood; and, secondly, because e urticaria and articular pains, which are to the patient sources of the ost serious discomfort, are largely avoidable incidents.

In connexion with this I showed some ten years ago that these incidents re associated with a diminution in the coagulability of the blood, pointing ut as I did so that this condition of diminished coagulability and viscidity tands in causal relation to "serous haemorrhages" generally, and

[1] *Die Serumkrankheit*, Franz Deuticke, Leipzig und Wien, 1905.

drawing attention to the fact that it is possible to restrain such serous haemorrhages by the administration of calcium salts.[1]

My suggestion that it would be possible by the exhibition of calcium salts to forestall the urticaria and articular troubles of the serum disease and to cure these rapidly when they have supervened, has recently been completely justified by the work of Netter.

Netter has shown in a very conclusive manner, by systematic observations conducted with controls on some hundreds of cases, that the percentage of urticarias in patients treated with diphtheria antitoxin can be very strikingly reduced—it was reduced, I think, in Dr. Netter's series of cases by four-fifths—by the exhibition of calcium chloride.

I would, therefore, again commend this procedure as a routine measure after serum injections, in particular in connexion with prophylactic inoculations of diphtheria antitoxin.

It would probably suffice in the case of an adult to administer 30 grains of calcium chloride or lactate daily from the sixth to the tenth day after the injection of serum.

Having disposed of this incidental and relatively speaking unimportant issue in connexion with serum therapeutics, let me now turn to the main theme of my discourse. In serum-therapy the physician proposes to administer to the patient protective substances elaborated in the organism of an animal which has been vicariously inoculated with the appropriate bacterial vaccine. The method is commended to the physician among others by the following considerations. He believes that, incidental troubles such as urticaria apart, he cannot by serum-therapy do any harm even if he fails to do any good. He believes that every serum which bears the label of a reputable laboratory, stating that it is an "antiserum" for a particular microbic infection is, as it professes to be, a serum which contains protective substances and a serum which contains these in sufficient quantity to reinforce to a sensible extent the antibacterial substances in his patient's blood. Further, he believes that in choosing his dose he may guide himself entirely by the clinical condition of his patient, employing more of the serum when the patient's condition is very serious, less when his symptoms are less urgent. And above all, the physician believes that he has behind him in the capacity of a scientific guardian a bacteriologist who, as a preliminary to issuing the serum, has arrived at an adequate knowledge of its composition.

The whole of this body of beliefs rests, I am convinced, upon a foundation of sand. Let me for the moment deal only with the last and most

[1] "On the treatment of the hæmorrhages and urticarias which are associated with deficient blood-coagulability."—*Lancet*, Jan. 18, 1896.

Notes on two cases of urticarias treated by calcium chloride.—*Brit. Journal of Dermatology*, vol. viii, No. 89, 1896.

On the association of serous hæmorrhages with conditions of defective blood-coagulability.—*Lancet*, Sept. 19, 1896.

In each of these papers cases of serum disease treated by calcium chloride are reported.

nportant belief, with the belief of the practitioner that the bacteriologist ho furnishes the serum has arrived at a competent knowledge of its omposition. The bacteriologist will emphatically repudiate any such nowledge in connexion with many of the sera which he furnishes. Ie will, no doubt, in the case of his antidiphtheria serum, and in the ase of his antitetanus serum, tell you the antitoxic value of the serum; r he will have measured it. But he will admit to you—if you press ome your inquiries—that in the case of antibacterial sera, where he as not at disposal any adequate methods of examination, his sera re often sent out without any searching examination. His only war-anty for labelling his sera in such a case as "antisera" is derived om the fact that he assumes that horses may be trusted to furnish otent antisera when injected with progressively increasing doses of ny bacterial vaccine.

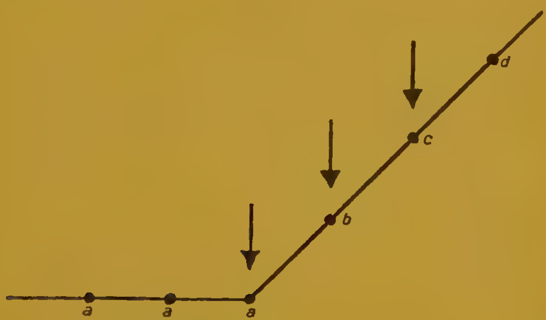


Fig. 1.

It is for you to consider whether this assumption of the serum manu-acturer is an assumption which he is entitled to make, and an assump-on upon which you are justified in reposing confidence.

Let us try to make clear to ourselves the working theory of the acteriologist who manufactures sera. He would probably formulate s working theory in some such form of words as the following: "If in a diagrammatic manner, represent the amount of protective sub-ances in the blood of a horse anterior to inoculation by a series of ots positioned as at *a*, *a*, *a* (Fig. 1), the amount of protective substances that horse's blood after recovery from a first inoculation of any acterial vaccine may be represented by a dot positioned as at *b*. After cond and third inoculations, conducted in each case with doses increas-g by geometrical progression, the amount of antibacterial substances ay be represented by dots positioned at *c* and *d*. Progressing in this anner, I shall arrive at a condition when there will be contained in e horse's blood very large amounts of protective substances."

When we come to inquire into the basis upon which this comfortable eory is built up, we find that results such as those just postulated are

obtained in connexion with the inoculation of diphtheria and tetanus toxins into horses. But let it be noted that results such as these are, even in connexion with the inoculation of diphtheria and tetanus toxins into horses, not always obtained. In a certain percentage of cases no satisfactory amount of antitoxin is ever achieved. Again, a certain number of horses succumb in the course of the treatment. Lastly, a condition may be arrived at in which injections of toxin are no longer responded to by an elaboration of antitoxin.

It will be recognized, in view of the above, that the generalization upon which the serum manufacturer relies is not a true generalization from the whole of his experience in connexion with diphtheria and tetanus antitoxins. It is a generalization from his experience in connexion with his successful cases.

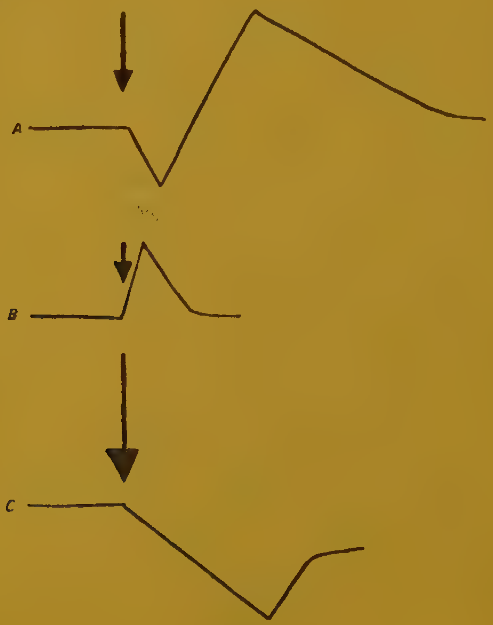


FIG. 2.

The serum manufacturer has thus not even the justification of an argument from analogy for assuming, as he does, that the serum which he obtains by the inoculation of bacterial cultures into horses must contain protective substances. And he has even less justification in assuming, apart from actual verification, that his sera contain protective substances in sufficient concentration to be therapeutically useful. In so fundamental a matter as this the proper course for the bacteriologist is, as you will recognize, to verify everything, to measure everything,

d to declare in each case the results of his measurements. In parti-ar ought the alterations in the anti-bacterial powers of the blood ich supervene upon the incorporation of bacterial vaccines into the rses which serve for the production of his sera in each case to be inves-ated. I take it as assured that those alterations would correspond all respects to those which I found to occur in man in connexion th antityphoid inoculation, and afterwards in connexion with the oculation of very many different varieties of bacterial vaccines. I may rhaps recall to your minds the sequence of events which occurs in each se, and which I have spoken of as "the law of the negative and positive ase," or as "the law of the ebb and flow and reflow of the wave of munity." Let me trace for you here a typical curve representing e changes in the antibacterial power of the blood which occur after an dinary inoculation (Fig. 2, A).

This form of the curve may be modified. Here, for instance (Fig. 2, B), ere only a small dose of vaccine is administered, the negative phase elided, and duration of the positive phase is shortened. Again, where large dose is inoculated the negative phase is accentuated, until finally e positive phase is abolished, the blood remaining for a considerable riod below the level at which it originally stood (Fig. 2, C).

The significance of these curves in connexion with serum therapeutics ll immediately come home to you. It will be appreciated that accor-ng as a larger or smaller dose of bacterial vaccine has been incorporated to the particular horse, and according as an earlier or later date may ve been chosen for the bleeding of that horse, his blood will contain, the one case, unneutralized bacterial poisons and less protective sub-ances than the normal blood, and in the other case protective sub-ances in excess of those found in the normal blood.

In the curves I have drawn (Fig. 2) I have set forth, as you will have served, only the events which supervene upon a single inoculation. Let e now, seeing that we have in serum-therapy to deal with sera which e obtained after a whole series of inoculations, pass on to consider th you the results which may be achieved by a series of inoculations.

Here one of three things may happen. (1) Each inoculation may erate as an independent event, as indicated in Fig. 3, A; (2) the inocu-tions may produce a cumulative effect in the direction of the negative ase, as indicated in Fig. 3, B; (3) the inoculations may, as in Fig. 3, C, oduce a cumulative effect in the direction of the positive phase.

The first result is, I believe, the ordinary result of inoculation of a cterial vaccine when the animals are allowed to recover completely tween the successive inoculations.

The second result is, I suspect, very frequently achieved when the oculations are pushed.

The last effect would, if I may argue from a very large experience the effects of inoculation on man, appear to be only exceptionally alized. Moreover, even where the very best results are achieved in con-

nexion with the elaboration of anti-bacterial substances in the organis[m] that miracle of successful immunisation which accomplishes itself in co[n]nexion with the inoculation of diphtheria toxin and tetanus toxin in horses—a miracle which gives us, often within the compass of a ve[ry] few cubic centimetres of serum, a quantum of antitoxin which suffices [to] alter the whole course of the patient's disease, is, so far as we know els[e]where, never even distantly approached.

You will recognize how different all this is from the dream which yo[u] share with the serum manufacturer. The serum manufacturer makes h[is] serum, and you administer it, thinking that beyond doubt you are admini[s]-

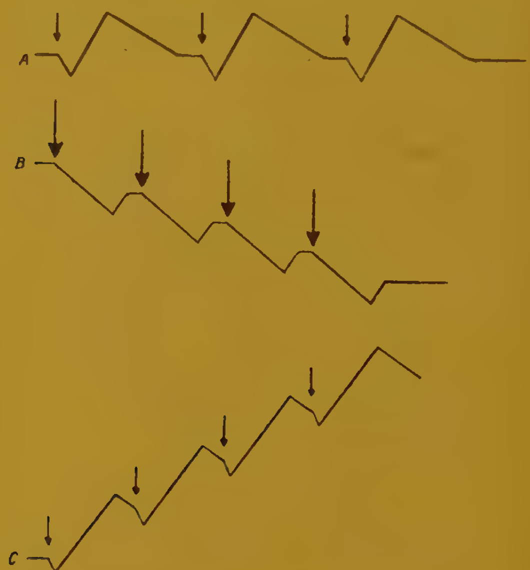


Fig. 3.

tering a quantum of antibacterial substances which will count in you[r] patient's conflict with his bacterial invasion. And all the while th[e] product which the serum manufacturer is sending out, and which you ar[e] administering, may not contain any appreciable quantity of protectiv[e] substances.

That serum may, on the contrary, contain, practically unaltere[d], the identical bacterial poisons which were originally incorporated int[o] the horse. And by the agency of these it may so happen that you may, as by the incorporation of a dose—here an unmeasured dose—of [a] bacterial vaccine, be lowering, temporarily at least, your patient's power[s] of resistance with respect to the micro-organisms with which he has t[o] deal.

In connexion with this last possibility, two questions immediatel[y]

ress for solution. The *first* is the question as to where the responsibility sts if a serum which contains a toxic bacterial element is administered as serum which contains protective substances. The *second* is the question to whether there are any stigmata by which the bacteriologist, or iling him, the clinician, would be capable of discriminating between a rum which contains protective substances and a serum which contains xic bacterial elements.

In particular, the issue as to where the responsibility rests in the case the administration of a serum which is something quite other from hat it is assumed to be is an issue which must come home to us all. We ay take it that the physician will contend that the responsibility lies the shoulders of the bacteriologist, and will point to the fact that e serum is labelled as an "antiserum" as a fact which places the cteriologist clearly in the wrong if the serum does not contain pro- ctive substances in appreciable quantity, and above all, if it contains xic bacterial elements.

The bacteriologist, for his part, will, no doubt, contend that when has incorporated into the horse his bacterial vaccines, and when he s waited after the last inoculation until the horse is "restored to alth," he has fully discharged his responsibility, and that the clinician ust be answerable for any results which may follow from the exhibition the serum.

It may be debatable as between physician and the bacteriologist as to ho is to take the blame, but it is for us to look at the matter from the tient's point of view. From the point of view of the patient someone assuredly to blame if a serum is administered which is quite other from hat it is intended to be, and assuredly, also, we must hold the practi- oner who administers the serum as responsible to the patient.

If I am correct in this, the physician should abstain from assuming ith regard to his patient responsibility for any serum except where, in the case of an antidiphtheria serum, the bacteriologist is prepared give him definite information with respect to the amount of protective bstances in the serum. Again, all experimentation with sera of uncer- in composition ought, I submit, to be undertaken exclusively by those ho, understanding the possibilities of error, would proceed in these mat- rs with a due sense of responsibility. I shall return to the subject later.

For the present, let me turn to the second of the issues spoken of ove, and let me consider with you whether there is any possibility determining in connexion with a serum which purports to be an anti- cterial serum whether it is rich in antibacterial elements, or whether it ntains toxic elements derived from the bacterial vaccines which were oculated in the horse.

There are in point of fact a whole series of methods which may be ploited in this connexion.

1. It may be determined upon animals whether a serum which rports to be an "antiserum" does or does not possess toxic properties.

In this connexion attention may be drawn to the fact that a pa ticular "antistaphylococcus serum" which was examined by D Bulloch was found to be extremely toxic.

2. It may be determined upon animals whether, failing the pr duction of an overt toxic effect on animals, the incorporation of a seru reduces the resisting power of the animal organism to the correspondi bacterial infection.

In this connexion attention may be drawn to the fact that one the "antiplague sera" which was furnished to the Indian Plague Co mission for trial upon man, and which fell to me as a member of th Commission to test, proved to be a serum which, while it was not definite toxic in the sense that the antistaphylococcus serum referred to abo was toxic, was none the less a serum which lowered the resisting pow of guinea-pigs to plague infection, causing these animals to succumb the test inoculations in shorter time than the control animals—t

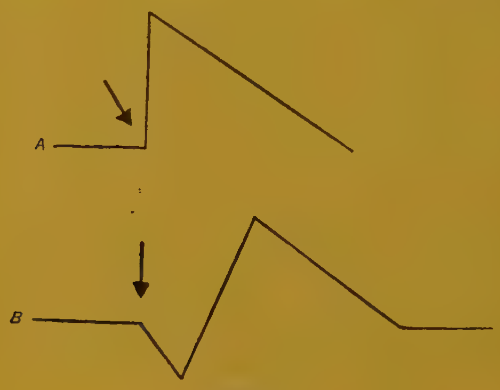


FIG. 4.

extent to which death was hastened being in each case directly propo tional to the dose administered.

3. It may be determined whether the addition of a serum to norm human serum does or does not deprive this serum of its antibacteri powers. I may here instance the fact that a highly agglutinating " ant typhoid serum" which was suggested for use upon the troops in Sout Africa, proved to be a serum which could not be added even in sma quantity to normal blood *in vitro* without abolishing the bactericid power which is normally exerted upon the typhoid bacillus by huma serum.

4. Lastly, a serum which purports to be an antibacterial seru may be tested by the method which finds illustration in the Curv above.

The curve which I have labelled A would, I take it, be accepted b everyone as the type of curve which we are entitled to expect when w

ake a series of measurements of the protective substances of an ani-al's blood in connexion with the intravenous injection of an anti-rum.

The curve which I have labelled B would, I take it, be conceded to e the type of curve which would be obtained in connexion with the in-avenous inoculation of a bacterial vaccine.

The inference would appear to be justified that where the latter instead the former type of curve is obtained in connexion with the inocula-on of a serum; that serum must contain, not antibacterial substances, it bacterial elements derived from the vaccine which was employed in e "immunisation procedures."

I have applied this test in connexion with a serum which was derived om a reputable laboratory, and which was labelled "antitubercular rum," with the result that I obtained in the case of a rabbit inoculated ith this serum a curve of the type B—a curve which was practically entical with that obtained in the case of a control rabbit inoculated travenously with tuberculin. I may point out to you that this obser-ation falls into line with the fact that typical "tuberculin reactions" ave over and over again been recorded in connexion with the exhibition "antitubercular sera" to tubercular patients. You will, in view this, I think, be with me when I urge that not alone such experiments the above on animals, but if possible also measurements of the effect erted on the antibacterial power of the human patient, ought pre-minary to its issue to be made in connexion with every serum issued r the treatment of septicæmic disease. Where such definite measure-ents of the effect exerted by a serum upon the blood of patients have ot been carried out, it is sometimes practicable to discern the character the serum from clinical observation alone.

1. The clinician would be justified in suspecting the presence of icterial poisons in a serum in those cases where a tuberculin reaction other similarly characteristic reaction supervened upon the exhibition the serum.

2. He would perhaps be justified in a similar suspicion where severe onstitutional symptoms other than those which are characteristic of e serum disease, supervene upon inoculation—in particular where vere symptoms of intoxication make their appearance with greater equency in connexion with the treatment of the graver cases than in onnexion with the less serious cases of the disease.

3. Lastly, the clinician would be justified in suspecting the presence bacterial elements in serum where there supervenes immediately upon s inoculation an exacerbation of patients' symptoms, followed after an terval by a distinct improvement.

This particular sequence of events would, I submit, find its most tisfactory explanation in the *hypothesis that the serum is functioning s a vaccine.*

The idea that a serum may in reality be a bacterial vaccine in dis-

guise, and the idea that benefit may be derived from the inoculation a bacterial vaccine in cases of septicæmia are new and as yet unventilat ideas. I am anxious to commend these ideas to your consideratio Let me begin by putting it to you once again that, in view of all th we know with respect to the difficulty of obtaining a cumulation antibacterial substances in the blood, and in view of what we kno with respect to the rule of thumb manner in which the immunisation horses is at present conducted, it is quite as likely that a residuum of t bacterial vaccines originally incorporated should be contained in the seru issued as that a really useful quantum of antibacterial substanc should be contained in it. If this is so, a serum obtained by th immunisation of horses, while it may at one time be a vehicle f antibacterial substances, may at other times quite well be in effect bacterial vaccine.

Passing to consider the possibility of bacterial vaccines renderin useful service in connexion with the treatment of septicæmic infection I have to confess that the idea that bacterial vaccines could here play useful rôle was only a short time ago very uncongenial to my precon ceived notions. I conceived that when bacteria found access to th blood and generalized themselves in the system the machinery for immur ization which is at the disposal of the organism was fully called int action. In accordance with this I assumed that to inoculate bacteri vaccines in such circumtances would be to add fuel to the fire without con tributing anything to the elaboration of those antibacterial element which serve to extinguish the conflagration.

I have now recognized that both the premises upon which I buil and my inferences from those premises are assailable.

In the first place, while it is true that the machinery for immuni sation with which the organism is provided is called into action i septicaemic diseases, it would seem certain that under the condition which obtain in such diseases that machinery is often not working t its full capacity.

An unmistakable indication of this is furnished by the fact that i the exactly analogous conditions which obtain where bacterial culture or the filtrates from these are inoculated intravenously into horses ther is obtained, in some cases at least, only a very poor yield of protective sub stances.

Again, in the reasoning which I above rehearsed, the possibility of different effect being produced by bacterial elements introduced into th blood stream and the same bacterial elements introduced directly int the tissues was overlooked. Yet consideration will show that ther may be quite important differences, first in the matter of the toxic effect exerted, and secondly with respect to the immunising response elicite by one and the same quantum of bacterial elements introduced directl into the blood stream, or, as the case may be, directly into the tissues The general intoxication effect—which above all we have to apprehen

n septicaemic conditions—may be expected to be greatest where, as ccurs in these infections, bacterial derivatives find direct access to the rculating blood, and least where, as would be the case in the inocula-on of a vaccine, the bacterial elements are introduced into the tissues. n this latter case, as may often be seen in connexion with the inocula-on of small quanta of antityphoid bacilli when the patient keeps his ed after the inoculation, the toxic effect of the vaccine may expend self exclusively upon the tissues at the seat of inoculation, constitu-onal symptoms being here practically absent.

Equally important are the differences which may manifest them-elves in the matter of the immunising response according as one and he same quantum of bacterial elements is incorporated into the blood, r, as the case may be, directly into the tissues. In the case where acteria are, as in septicaemic conditions, found in the blood stream, r in organs standing in direct relation with this, the bacterial derivatives re of necessity diluted by the whole volume of the blood and lymph efore they can come into application upon the tissues in which, we nay take it, the machinery for the elaboration of protective substances s located. In conformity with this great dilution of the bacterial erivatives a comparatively speaking ineffective immunising stimulus vill here be administered. In contrast with this, where a bacterial vaccine is inoculated directly into the tissues, the bacterial products vill come into application upon these in a very concentrated form, alling forth a correspondingly larger production of protective sub-tances.

Such larger production of protective substances is, in point of fact, egularly achieved in the horse in connexion with the production of liphtheria antitoxin, when, in lieu of intravenous inoculations, subcutane-ous and intra-muscular inoculations are resorted to, and, it would seem, in particular in the case where the inoculations are made with very con-centrated toxins. My fellow-workers and I have also recently, in con-nexion with the treatment of a considerable number of cases of Malta fever, and in connexion with one case of infective endocarditis—the details of which will be elsewhere published—achieved by the exploita-tion of the corresponding vaccines in carefully controlled doses both an ncreased production of protective substances, and in each case in association with this a rapid cure.

Having now, I hope, made it clear that a serum which is adminis-tered as an "antiserum" may in reality be a bacterial vaccine in disguise, and having further indicated to you that bacterial vaccines may be exploited with very happy results in connexion with the treatment of septicaemic disease, I think that you will bear with me when I suggest to you that many of the successful results which have been achieved by serum-therapy would find their simplest explanation in the assumption that the sera which were exploited operated as bacterial vaccines.

The idea that this might be so suggested itself to me first in co nexion with a particular sample of highly agglutinating anti-Malta fev serum which was prepared and issued by me from the Pathologic Department of the Army Medical School, Netley. It was reporte to me with respect to this sample of serum that it produced in the tw Malta fever patients upon whom it was employed in each case, as i first result, an alarming exacerbation of the fever, and in associatio with this in each case very severe urticaria and oedema. After the laps of a few days there supervened a sudden turn for the better, resultin I understand, in each case in a rapid cure.

Looking back on these cases, and calling to mind on the one han the fact that this sample of serum was obtained only after the hors had been subjected to a long course of what was throughout in inter tion a process of immunisation; and on the other hand, the fact tha nothing at all resenbling the effects which were obtained by this sampl of serum had been obtained with samples drawn off earlier in the cours of the horse's "immunisation," which were tested upon Malta feve patients—among others upon myself, at a time when I came under tha category—and correlating with these facts the fact that an exacerbatio of the fever is seen also in connexion with the inoculation of a Malt fever vaccine into Malta fever patients, I regard it as in the highes degree probable that the serum which caused the effects described abov must have contained toxic elements derived from the Malta fever vac cines which had been incorporated into the horse.

I have seen a train of events somewhat similar to that recounte above supervene also in connexion with the inoculation of antistrepto coccus serum, and I suspect that some of you may also have had simila experiences, for I gather from the literature that intoxication symptom followed by improvement in the patient's condition are from time t time met with in connexion with the inoculation of many differen "antisera."

Isolated facts cannot, however, furnish a basis broad enough fo generalization such as that which I am seeking to commend to you acceptance. You will, I doubt not, as a precondition to embarking upon any serious consideration of the hypothesis I have suggetsed to you, desire to be furnished with a summary of the best results which have been obtained in connexion with the serum-therapy of septicaemic diseases; and you will desire to have furnished to you, in connexion with that summary, data which would countenance the idea that the results obtained may properly be referred, not to protective substances, but to toxic bacterial derivatives contained in the serum employed.

It so happens that in a paper recently published by Chantemesse on the treatment of typhoid fever by an "antityphoid serum" I find as nearly as possible all that I require for compliance with your wishes. Let me place you in possession of the facts which are recorded in Professor Chantemesse's paper. He has during the last five years treated

ch his "antityphoid serum"—if I understand aright—all the cases, 2 in number, which have been admitted to his typhoid wards. By acing side by side with the results obtained upon these patients the sults obtained without serum in the typhoid wards of all the other aris hospitals taken together, the following comparison is obtained :

	Number of Cases of Typhoid Fever treated.	Number of Deaths which occurred among these.	Case Mortality.
			Per cent.
eated without serum . .	3,595	753	17·3*
eated with serum . . .	712	27	3·7

* M. Chantemesse's arithmetic would here appear to be at fault.

It cannot be doubtful to any one who considers these results, or ho has had the privilege, as I have had, of visiting Prof. Chantemesse's ards and seeing his cases, that we have here a very brilliant and, I esume, an absolutely unique achievement in connexion with the eatment of typhoid fever.

Our present concern in connexion with that achievement is to conder whether the results which have been obtained are to be imputed protective substances elaborated in the organism of the horse and ansferred to the patient in the vehicle of the serum, or to a residuum f the typhoid vaccine originally inoculated into the horse and transrred to the patient in the vehicle of the serum.

This fundamentally important issue is not discussed in M. Chanteesse's paper. We are, however, supplied with certain data from hich we can make our own deductions.

Let me try to marshal these data for you :

1. We learn in M. Chantemesse's paper that two well-marked phases ome under observation in connexion with the inoculation of the serum. mmediately after inoculation there supervenes a "phase of reaction." his phase may last from four to five days. It is associated with an xacerbation of the patient's symptoms.

Upon this phase of reaction follows a "phase of defervescence."

There would seem to me to be little doubt that these two phases must e identified with the "negative" and "positive phases" considered arlier in this paper. In other words, it seems to me clearly indicated ere that the inoculation of the serum is followed by a reduction in the ntibacterial power of the blood—a reduction which stands in relation to he exacerbation of the fever—and that this is followed up by an increase n the antibacterial power of the blood, standing in relation with the defervescence which is observed.

2. We learn that the dose of the serum which is employed is very small—"a fraction of a cubic centimetre"; further, that this dose is reduced proportionally with the severity of the symptoms and the gravity

of the case. It may even, I understand, be reduced in very severe ca to "half a drop."

Again, we learn that when reinoculation is resorted to the dose further cut down by one half; and, lastly, we learn that where reino lation is undertaken it is undertaken only after the expiration of a co siderable number of days—in the case of the published curves aft twenty-three and sixteen days respectively.

When we come to ask ourselves whether this scheme of dosage, scheme of dosage which has given the brilliant results above recorde can be reconciled with the idea that we are dealing here with a tr "antityphoid" serum, it becomes clear as noonday that the scheme dosage employed is the exact reverse of that which would be apposite if were dealing with a true "antiserum" such as antidiphtheritic serum

When dealing with a true antiserum one does not limit the do in an ordinary case to "a fraction of a cubic centimetre," nor does o diminish the dose to "half a drop" in the gravest cases—those cas where the organism stands most in need of protective substances. Rath does one, where a true antiserum is to hand, give in each case, unle where limited by financial considerations or considerations which ha reference to the serum disease, large doses of serum. Again, in the mo serious cases one lets all the considerations above referred to go by th board and gives larger and larger doses. And, yet again, where th result which one desires is not achieved by a single inoculation one do not allow a long interval to elapse before reinoculation.

In contrast to all this, when one is exploiting a bacterial vaccine i the treatment of a septicaemic disease one would, except for the fac that the dose can much more appropriately be regulated by the measure ment of the antibacterial power of the patient's blood, follow a schem of dosage which would be an exact replica of that followed by M Chantemesse. I think I need not further labour my point.

3. Let me turn to a further series of considerations which sugges that the antityphoid serum which M. Chantemesse has so brilliantl exploited is a typhoid vaccine in disguise.

It is, as all who have attempted it know, a very difficult task in the cas of the horse to carry immunisation against the typhoid bacillus up to th point at which a satisfactory "antiserum" is produced. There is thus a *à priori* improbability that M. Chantemesse has obtained in his serum a therapeutically useful quantum of protective substances. Nor doe M. Chantemesse, apart from the fact that his serum is efficacious i reducing the death-rate in typhoid fever, claim very much for his serum What he claims for it amounts to no more than this—that the serum possesses a considerable agglutinating power, and that a quantum o serum corresponding to the maximum dose which is employed thera- peutically upon man—that dose, if I understand M. Chantemesse aright never exceeds "a fraction of a cubic centimetre"—may with impunity be inoculated intra-cerebrally into rabbits and guinea-pigs.

I would ask you to note that neither the one nor the other of these ıdings is in any way inconsistent with the idea that the serum may ve its efficacy to the presence of typhoid vaccine.

The presence of agglutinating elements in the serum cannot, as we ıve seen, be accepted as evidence of the absence of bacterial derivaves. You will remember that the toxic "anti-Malta fever serum" hich was discussed above was possessed of well-marked agglutinatg power. And you will remember also that the "antityphoid rum," referred to above as a serum which completely abolished the ıctericidal power which the normal blood exerts upon the typhoid ıcillus, was in like manner a serum which possessed a well-marked— deed, an altogether exceptionally well-marked—agglutinating power.

If the presence of agglutinins cannot be accepted as evidence of the eedom of a serum from toxic properties, much less can in the case of ı "antityphoid serum" experimental results such as those obtained y M. Chantemesse on rabbits and guinea-pigs be accepted as adequate vidence. These I regard as quite inconclusive, first, because very ıarked constitutional symptoms may be produced in man by doses of yphoid vaccine which are insignificantly small in comparison with ıose which are required to kill rabbits or guinea-pigs; and secondly, ecause, as pointed out earlier in this paper, the resistance of the organism ɔ bacterial invasion may be seriously reduced by the exhibition of the erum, even where ostensible symptoms of poisoning are completely bsent.

Scrutinizing M. Chantemesse's successful results from all the different oints of view which I have asked you to consider, I think there is no vay of escape from the conclusion that his results—and I would urge hat these are the only statistical data which are conclusive as to the tility of serum-therapy in connexion with septicaemic disease—are ı all probability to be regarded as results obtained by the inoculation f bacterial vaccines.

Let me try now to bring out in clear relief the salient points I have ndeavoured to establish in this discourse.

1. I have tried to make good to you the proposition that the sera vhich are issued as "antisera" for the treatment of septicaemic conlitions may, as the case may happen, contain a certain quantum—not ıecessarily a therapeutically useful quantum—of protective substances laborated by the horse, or they may be inert, or finally they may contain bacterial elements derived from the culture originally inoculated nto the horse.

Any one of these three varieties of serum may at present be sold to you under the denomination of an "anti-serum."

2. I have drawn your attention to the fact that the scheme of dosage which would be indicated in the case of a serum which contained only protective substances would, if followed in the case of a serum such as was last in question, be dangerously erroneous. In like manner a

scheme of dosage such as would be appropriate in the case where t serum is the equivalent of a bacterial vaccine would, in connexion w a true antiserum, be absolutely ineffective.

3. Lastly, I have endeavoured to bring home to you that th is good reason to believe that the only brilliant results which have be achieved by serum therapy on a series of cases—to wit, the results M. Chantemesse—have been achieved by the inoculation of a seru which may be presumed to have functioned as a bacterial vaccine.[1]

If useful serum-therapy in septicaemic diseases resolves itself into question of the inoculation of bacterial derivatives, there is no need, think, for me to point out to you the moral.

[1] *It appears from his paper read before the International Congress for Hygie and Demography, Berlin,* 1907, *that M. Chantemesse has now tacitly adopted the vi which is here put forward.*

It would seem clear also from many of the descriptions of the clinical sympto (negative and positive phase effects) which have been observed after the injection of Ma morek's "antitubercular serum" that we have here also to deal with an "active i munisation" insinuating itself under the mask of a "passive immunisation."

Synopsis of the Principles of Vaccine-Therapy and Therapeutic Immunisation Generally.[1]

Being the Substance of a Series of three Herter Lectures delivered before the Johns Hopkins University, Baltimore, and of a Lecture delivered before the Harvey Society of New York, in October, 1906.

By A. E. WRIGHT.

I. Introductory Criticism.—(1) Treatment by Chemical Antiseptics—(2) Treatment by the Extirpation of the Obtrusive Focus of Infection—(3) Treatment by the Determination of Lymph to Focus of Infection—(4) Serum-therapy—(5) Expectant Treatment.

II. Principles of the Method of Vaccine-therapy, and Sketch of the Machinery of Immunisation?

Question as to how far the Particular Composition of the Vaccine is a Material Element in the Success of an Inoculation Process.

III. Question as to what is the Best Method of Gauging the Immunising Effect of a Vaccination—(1) Test Inoculations with Living Cultures undertaken upon Vaccinated Men or Animals—(2) Test Inoculations with Living Cultures undertaken, not upon the Vaccinated Organism, but Vicariously upon Animals treated with Serum derived from the Vaccinated Animal or Man—(3) Consideration of the direct Toxic Effects produced by Inoculation.—(4) Consideration of the Improvement or Aggravation of the Clinical Condition which follows upon Inoculation—(5) Evaluation of the Antibacterial Power of a Sample of the Patient's Blood; and Discussion of Question as to whether we ought to aim at a "Complete" or at a "Partial Evaluation"—Grounds for taking as our Routine-guide in connexion with Immunisation Procedures the Estimation of the Opsonic Index.

IV. Detailed Study of the Curves of Immunisation which are obtained by the Inoculation of Bacterial Vaccines—Question as to whether it would not be possible, by piling one Inoculation upon another, to achieve a larger Output of Antibacterial Substances, and in Association with this a Positive Phase of Longer Duration—Question as to whether it is expedient to employ in Successive Inoculations Progressively Increasing Doses of Vaccine, and Suggestions with Respect to the Regulation of the Dosage—Question as to the Site which ought to be selected for the Inoculation of a Bacterial Vaccine.

V. In addition to mastering the Physiology of the Immunising Response the Immunisator must concern himself also with the following:—

(i.) Auto-inoculations—Assistance which can be obtained from Purposely Induced Auto-inoculations in the Diagnosis of Obscure Cases of Localized Bacterial Infection.

(ii.) Conditions which obtain in the Foci of Bacterial Infection.

VI. On the two Broad Principles which ought to guide us where we set ourselves to combat Bacterial Infections by the agency of the Protective Elements which are furnished by the Organism.

(i.) Comparison between the Therapeutic Policy which is embodied in these Principles, and the Policies embodied in the Measures which are currently employed in the Treatment of Bacterial Infections—Rationale of the Policy which is embodied in the two Therapeutic Principles formulated above.—Question as to which of these two Therapeutic Principles is to be followed in those Cases where they come into conflict.

[1] Originally published in the *Lancet*, August 17 and 24, 1907. *Revised and brought up to date, Oct.* 31, 1908.

I.—Introductory Criticism.

GENTLEMEN,—I have undertaken to outline to you—so far as I can do
within the limits at my disposal—the principles of vaccine-therapy-
i.e., of the treatment of bacterial disease by the inoculation of the corr
sponding vaccines.

Let me preface what I have to say to you on this subject by askin
you briefly to review with me the methods—other than vaccine-therap
—which we have to-day at disposal for the treatment of bacterial diseas

The following are, I think, the only methods which come here int
consideration: (1) *treatment by chemical antiseptics;* (2) *treatme
by the extirpation of the obtrusive focus of infection;* (3) *treatme
by the determination of lymph to the focus of infection;* (4) *serum
therapy; and* (5) *the "expectant treatment."*

(1) Treatment by Chemical Antiseptics.

Antiseptics have found in medicine a threefold application. The
have been locally applied with a view to holding in check and extinguish
ing localized bacterial infections. They have been used for the purpos
of checking putrefaction in discharges and devitalized tissues. The
have been administered internally with a view to checking microbia
growth in the blood, or in regions which can be reached only by th
channel of the blood.

Neither the second nor the third of these applications calls for any
discussion. There has never been any doubt as to the possibility o
suppressing putrefactive changes by antiseptic irrigations. And i
is now all but universally recognized that it is futile to attempt to check

[1] We have a general formula for such antiseptics when, in technical terms
which perhaps improve a little on those suggested by Ehrlich, we say that they
are more "*histotropic*" than "*parasitotropic.*"

acterial growth in the interior of the organism by antiseptics [1] which ave—as our present antiseptics have—a greater affinity for the con- tituent elements of the body than they have for any bacteria.

Attention may therefore here be concentrated upon the issue as to hether the antiseptic applications are effective in holding in check nd extinguishing localized bacterial infections.

It is, of course, currently believed that this method of treatment s effective. It is in this faith that the surgeon introduces antiseptics nto septic wounds or, where he happens to be so minded, into abscess avities. It is in this faith that the physician resorts in the case of ulmonary infections to antiseptic inhalations. And it is in this faith hat the dermatologist, gynæcologist, laryngologist, aurist, and genito- rinary specialist are strenuous—each in the application of antiseptics o the particular province of the body which he takes under his care. It will be profitable for us to collate the facts and to consider whether here is in reality any trustworthy basis for the belief that inspires all his practice.

Significant in this connexion appears to me the fact that antiseptics re now by general consent abandoned in connexion with the treatment f ordinary surgical wounds. Significant also is it that the practice of ntroducing antiseptics into abscess cavities—erstwhile so common— s now less and less frequently resorted to. Significant, again, is it that reatment by antiseptics in case of bacterial invasions of mucous mem- ranes is to-day more and more frequently followed up by curetting, craping, and so-called "radical" operations. Above all significant, s it that so distinguished a dermatologist as Sabouraud should sum up he results of antiseptic treatment of bacterial diseases of the skin as ollows: *ç'a été une chose curieuse que la faillite des antiseptiques dans le raitement des maladies dermatologiques parasitaires. On fondait sur ux des espérances colossales, ils n'ont presque rien donné.*"

The results which have been obtained in connexion with pulmonary nfections by antiseptic inhalations and in connexion with bacterial nfections of the genito-urinary passages by "urinary" and other anti- eptics are, I am persuaded, neither better nor worse than those which ave been obtained in connexion with diseases of the skin.

Now all this failure of antiseptics is, I submit, only what might *à priori* ave been expected. Let me put the case to you as I see it.

It is, of course, axiomatic that antiseptics can take effect only upon hose bacteria with which they come into contact. It is obvious also n the case of bacterial infections of the skin and mucous membranes hat the infecting bacteria will not all be lying on the surface, and that hey will not, when lying on the surface, be limited to those regions which re accessible to antiseptics. It follows that it will be quite unreasonable o expect from any application of antiseptics a complete sterilization. In every case a residue of bacteria will survive. Inevitably these will nultiply and reoccupy the disinfected surface.

[1] *Bulletin de l'Institut Pasteur*, 1904, p. 286.

And this is not all. The antiseptic will not, as the unthought assume, add its antibacterial power to the antibacterial power of t living organism. On the contrary, the antiseptic will directly antagon the protective forces which the living organism has at command. will paralyse the phagocytes, and will abolish the antibacterial power the blood fluids. The disinfected surface will thus, by the action the antiseptic, be left "swept and garnished"—ready for reinvasi by the expropriated bacteria.

And, again, this is not all. The antiseptic application will also inju the histological elements and, in particular, the capillaries of the tiss to which it is applied. It will thus lead to an outpouring of lymph fro the disinfected surface. That outpouring will not only wash away t antiseptic, but it will, where a skin surface is in question, convert t natural armour of the normal epidermis into a lymph-sodden pulp which bacteria will easily establish themselves.

(2) Treatment by the Extirpation of the Obtrusive Focus of Infection

Where the invading bacteria have penetrated into the interior of t body their destruction by antiseptic applications is recognized to be out question and the policy of proceeding against them by surgical metho comes up for consideration.

I can understand that extirpation may be imperative where an orga has been completely disorganized by invading bacteria and where the is danger to life from the spread of the infection. I can also understan that a case can be made out for extirpation where there is prospe of removing all the infecting bacteria without danger or sensible mutil tion. Lastly, I can understand that it will be an added advantage an extirpation operation removes along with the infecting bacteria useless organ which is specially subject to infection.

But assuredly these are not the conditions under which the majori of scraping and extirpating operations are undertaken. And in particula these are not the conditions which confront us in those extirpation opera tions which are employed as a routine treatment in connexion wit localized tuberculous infection. Here long before the surgeon has bee called upon the scene not a few of the bacteria may, by the agency the blood and lymph stream, have been carried beyond reach of his knif

It will be clear that it cannot be claimed for extirpation operation undertaken in these circumstances that they are in any real sense the term "radical operations."

They are operations which aim only at the extirpation of one or mor obtrusive foci of infection.

In harmony with this conclusion is, I submit, the fact that tuberculou disease so often recurs after operation—exceptionally, in the form general tuberculosis; commonly, as a localized process in the site the operation, or elsewhere.

Such results are not, I think, adequately explained by mechanical dis turbance in the focus of infection, or by the incomplete character

ne operation. There is probably at work here also another factor. . reduction in the antibacterial power of the blood may, as we shall see, apervene upon operative interference.

(3) Treatment by the Determination of Lymph to Focus of Infection.

The method of extirpation by the knife is only one of the methods hich can be employed for the treatment of bacterial infections when ese have passed beyond the reach of antiseptic applications.

Of the other methods the more important are the application of hot mentations, massage, the method of Bier, the various forms of radio-ierapy, free incision into infiltrated tissues, and the evacuation and rainage of abscess cavities.

In all of these methods—and it is this which has induced me to ring them together here under a single heading—we have a determina-on of lymph from the circulating blood to the focus of infection. Ve have also, except in those cases where an external vent is pro-ided, a conveyance into the circulating blood of a lymph which has passing through the focus of infection become impregnated with acterial products.

I shall, before I have done, explain to you, in connexion with the passage f blood fluids into the focus of infection, that these blood fluids will xert in every case some antibacterial effect upon the invading bacteria nd, in connexion with the passage of bacterial products from the focus f infection into the circulation, that these products will effect very nportant modifications in the blood both in the direction of reducing nd in the direction of increasing its antibacterial power.

We may, accordingly, from the passage of blood fluids into the focus of fection, expect in every case a certain therapeutic effect.

From dissemination of living bacteria, and from the conveyance of acterial products from the focus of infection into the blood which will ccur where no external vent for these is provided, we must, on the other and, expect occasional disaster.

You know that disaster has occasionally supervened upon massage, pon the application of Bier's bandages and upon radio-therapy.

(4) Serum-therapy.

All the therapeutic methods which have been in question above have pplication only to localized bacterial invasions. Serum-therapy has ore ambitious aims.

After a successful application in connexion with the treatment of iphtheria this method has—and this without intermediary trial in onnexion with simple, localized, and correspondingly more tractable rms of infection—been applied to the treatment of the most desperate nd complicated varieties of bacterial infection. Serum-therapy has, s you know, found application not only in connexion with septicæmic fections but also in those most complex mixed infections which we ave to deal with in pulmonary phthisis.

In these applications serum-therapy has, I submit, everywh disappointed expectation.

It is not enough to realize the fact of failure. Wherever, in co nexion with the application of any therapeutic method, we meet w repeated failure we are called upon to make a critical regress upon t facts and inquire into the rationale of that method.

It is exacted from us that we should, in accordance with this prin ple, here consider whether there is any assured basis for the treatme of bacterial infections by serum-therapy.

The serum-therapy of bacterial infection is, in point of fact, bu upon the postulate that the animal organism possesses the capacity responding to the incorporation of practically unlimited quanta bacterial cultures by a practically unlimited output of antibacter substances. Now this assumption, while it might be thought to w support from the analogy of the effective immunising response whi occurs where diphtheria or tetanus toxin is incorporated into hors is, as I think you will presently appreciate, entirely out of accord wi the results which are ordinarily obtained by the inoculation of bacter cultures. When you shall have considered how different are the ordina everyday events which supervene upon bacterial inoculations fro that miracle of immunisation which accomplishes itself when dip theria toxin is administered to a horse, you will, I believe, be quite pr pared to recognize that the *à priori* assumption that there will resu from the introduction of a moderate quantum of an inoculated anima serum into a patient's blood anything in the nature of a marked increa of his antibacterial power is entirely unwarranted.

(5) Expectant Treatment.

I now come to the expectant method of treating bacterial diseas the last of the five therapeutic methods which were enumerated the outset of this paper. We have, as you appreciate, in the expecta treatment a therapeutic method which is based upon the adoption a policy of non-intervention as between the invaded organism and t invading bacteria. We have here, in fact, a therapeutic method whic commits the destiny of the patient—so far as that destiny is involved the issue of his conflict with the invading microbes—entirely into t hands of chance.

It is agreed that by this method—which consists in little more tha feeding and nursing the patient and keeping him at rest in bed—far bett results are achieved in generalized bacterial infections than any th have been obtained by active medication. We need only to call to min here the fact that from 80 to 90 per cent. of recoveries are under the e pectant treatment achieved in typhoid fever.

There is, however, also a reverse to the medal.

We find there that we have in typhoid fever from 10 to 20 per cen of fatal cases, that we have in streptococcal septicæmias and in plague-

;o mention only two out of the more formidable bacterial infections—nortalities in comparison with which the percentage of recoveries is quite insignificant, that we have in Malta fever a considerable percentage of cases in which the fever drags out almost indefinitely—in short, that we have under the expectant treatment of septicæmic diseases in many cases a formidable percentage of failures.

This indictment of failure—of failure in connexion with septicæmic diseases—is very far from being the only indictment which can be brought against the expectant treatment.

Let me ask you to realize that generalized bacterial infections are, comparatively speaking, rare and transient incidents in life. The really serious ills of life are the various localized bacterial infections which sooner or later fasten upon every man, never afterwards releasing their hold.

It follows that it is a much less serious matter to say of a method of treating bacterial disease that it disappoints in connexion with many varieties of septicaemic disease, than it is to say that it has no application in connexion with localized bacterial infections.

Now the expectant method of treatment has no application in connexion with localized bacterial diseases.

It is only, if I rightly apprehend the matter, in the case where life is threatened by the entrance of bacteria or bacterial products into the blood—and, as we shall see later, not even invariably in that case—that Nature addresses herself in a serious manner to the task of immunisation.

As long as a bacterial invasion is still strictly localized it is idle to wait upon Nature and to expect from her any work of immunisation.

I am wont to insist in this connexion that the statistics of the expectant method of treatment are in the case of localized bacterial infections hardly more favourable than those of the Pool of Bethesda. You will remember, perhaps, with respect to that Pool, that an Angel was wont to come down and trouble its waters—was it *once* in seven years ?—and then—*only* the man who stepped down *first* into those waters was cured.

If it seems to you Nature must surely be kinder than this, I would ask you to take any series of cases of endometritis, middle-ear disease, chronic bronchitis, lupus, tuberculous caries, or let it be any other localized bacterial disease, and inquire in each case throughout what length of years the infection has persisted.

Among my own patients there are no fewer than three who have suffered from lupus for over 40 years.

II.—Principles of the Method of Vaccine-therapy and Sketch of the Machinery of Immunisation.

Having now passed in review with you the therapeutic methods which are in use in the treatment of bacterial disease, I propose to turn to the

main theme of my discourse and to deal with the treatment of bacteri
disease by vaccine-therapy. The essential feature of this method is th
scientific exploitation for therapeutic uses of the protective machiner
with which the organism is equipped. We shall do well to turn ou
attention here to a study of that machinery, remembering that no on
recovers from any bacterial disease unless it be by the production o
protective substances in his organism; that no one acquires protectio
against a disease except again by the production of protective substance
and that no one can live in the presence of infection except by the aid o
the protective elements in his blood.

There are two elements which come into consideration in connexio
with the protection of the organism against invading micro-organism
The leucocytes with their digestive ferments constitute one of thes
elements. The antibacterial substances in the blood fluids constitut
the other. A word or two will be appropriate in connexion with eac
of these elements of our protective machinery.

(*a*) *Leucocytes.*—The leucocytes come into consideration in connexio
with resistance to bacterial infection by virtue of the fact that they ar
capable of ingesting bacteria and of disintegrating these by intracellula
digestion.

We may usefully distinguish between "*spontaneous*" and "*induced*"
phagocytosis. By the former of these terms we may denote that proces
of ingestion which comes under observation when bacteria which hav
not been subjected to the action of the blood fluids are brought in contac
with washed leucocytes in an indifferent medium such as physiologica
salt solution.

Spontaneous phagocytosis, thus defined, is distinguished by the fact tha
it is a comparatively slow process; further, by the fact that the numbe
of bacteria ingested by each leucocyte attains ordinarily only very modes
proportions; again, by the fact that the ingestion is irregular in th
sense that individual polynuclear leucocytes differ very strikingly fron
their congeners with respect to their intake of bacteria—the majorit
picking up from such a bacterial suspension as is ordinarily employe
very few, if any, bacteria, while others ingest relatively considerabl
numbers; and lastly, by the fact that the ingestion of bacteria can b
completely suppressed by employing in the phagocytic mixture certai
concentrations—in the case of the tubercle bacillus a concentratio
of slightly over 1 per cent—of NaCl.

Strikingly different from such "spontaneous phagocytosis" is th
induced phagocytosis which comes under observation when leucocyte
are brought in contact with bacteria which have been, or actually ar
at the moment, subjected to the action of serum. The "induced phago-
cytosis" which occurs in these conditions is distinguished, first, by th
fact that it is an exceedingly rapid process; secondly, by the fact tha
every adult leucocyte, with hardly an exception, is here phagocyti
(instead of some few leucocytes taking part immoderately, while the other

abstain); thirdly, by the fact that the leucocytes will, in the case where the supply of micro-organisms is unrestricted, ordinarily fill themselves to absolute repletion; and fourthly, by the fact that the leucocytes will continue to ingest bacteria in a concentration of salt which entirely suppresses "spontaneous phagocytosis."

Seeing that phagocytosis of bacteria without subsequent intracellular digestion would, from the point of view of the protection of the organism, be meaningless, it will be plain to you that the digestive powers of the leucocytes would logically here come up for consideration. Into such discussion I am, however, for the present debarred from entering, not having as yet qualified myself to speak at first hand on the matter. Permit me, none the less, to give here my tribute of admiration to the brilliant initiating work which has been done on this subject by your fellow-countryman, Dr. Opie.

(*b*) *Antibacterial elements of the blood fluids.*—The blood fluids differ essentially from the nutrient fluids which are ordinarily employed by the bacteriologist for the cultivation of micro-organisms in the respect that, while the latter are nutrient fluids pure and simple, the blood fluids contain in addition to nutrient constituents also antibacterial elements.

The antibacterial elements which are here in question are "bacteriotropic elements" in the sense that they turn towards and enter into combination with elements of the bacterial body. Our knowledge of the modifications which are effected in the bacterial body under the influence of the bacteriotropic substances in the blood fluids is, it cannot be doubted, extremely incomplete. So much, however, already stands fast that the effect of the blood fluids upon the bacterial body may manifest itself in different ways.

The bacteria may be killed without being dissolved (*bactericidal effect*).

The bacteria may not only be killed but dissolved (*bacteriolytic effect*).

The bacteria may be so altered as to agglutinate in the presence of salt (*agglutination effect*).

The bacteria may be so altered as to be readily ingested by phagocytes (*opsōnic*[1] *effect*).

Inasmuch as the blood fluids produce in bacteria the different chemico-physical effects here enumerated, and inasmuch as agglutinating and opsonic effects can be obtained independently of each other, and independently of any bactericidal and bacteriolytic effects, it is convenient to assume that we have in the blood fluids four classes of bacteriotropic substances: *bactericidins*, *bacteriolysins*, *agglutinins* and *opsonins*.

Of these four varieties of bacteriotropic substances the opsonins would appear to be the most important.

We may ascribe to them a predominating importance, first, because it can be shown that the opsonic effect is, by either the normal or immune

[1] Derived from the Greek *ὀψωνέω*, and the Latin *opsōno*,—*I convert into palatable pabulum.*

blood, exerted upon every species of bacteria, whereas the agglutinatin effect is exerted only upon special varieties of bacteria, and the bactericida and bacteriolytic effect among pathogenic micro-organisms apparentl only upon the typhoid bacillus and the cholera vibrio.

The opsonins derive further practical importance from the fac that they can be very accurately measured (the error of estimatio in the case of ordinary bloods and in the hands of a good worker bein rarely greater than plus or minus 5 per cent.) and that it is possible seeing that the opsonic effect of the normal blood fluids is very marked to register not only (as in the case of the agglutinating power) an increase but also a reduction in the opsonic power of the blood.[1]

From the consideration of the protective machinery of the organism let me now turn to the consideration of certain preliminary points in connexion with the bacterial vaccines which operate upon that machinery. The first question which confronts us here is the question as to how fa the success of an inoculation process is dependent upon the particular composition of the vaccine. I have in view here in speaking of the particular composition of the vaccine not the axiomatic requirement that it must be affiliated to, or connected by affinity with, the microbes which it is designed to combat, but rather the minute details with regard to its source and manufacture.

Question as to How Far the Particular Composition of the Vaccine is a Material Element in the Success of an Inoculation Process.

Reflection will make clear that the success of an immunisation process must depend, in the first place, upon the power of immunising response which the organism may happen to possess with respect to the particular bacterial infection or intoxication process which is in question, and in the second place, upon (*a*) the composition of the vaccine and (*b*) the dosage and method of administration. Up to the present the composition of the vaccine has been considered to the practical exclusion of the question of dosage. Whenever ill-success has attended a process of immunisation, or whenever it has appeared to any one that it "ought" to be possible to improve upon the result already obtained; straightway the suggestion has been put forward that some change "ought" to be made in the composition of the vaccine. In such cases living vaccines have been proposed as substitutes for sterilized vaccines; vaccines derived from virulent cultures as substitutes for vaccines derived from avirulent cultures; vaccines derived from cultures more closely affiliated to the microbes against which protection is sought as substitutes for vaccines obtained from cultures less closely affiliated; vaccines derived from agar cultures for vaccines obtained from broth cultures; and

[1] Increase and reduction in the opsonic power of the blood is measured by comparing the amount of purely induced phagocytosis which is obtained with a normal blood with the amount of purely induced phagocytosis which is obtained with the blood of the patient under examination.

ιccines obtained by the trituration of bacteria, by autolysis, or treat- .ent by caustic alkalies and subsequent precipitation with acids, as ıbstitutes for vaccines which have been sterilized in the ordinary way y heat.

It has been claimed in connexion with each of the modifications ere in question that its employment is dictated both by weighty *à riori* considerations and analogies and by the event of actual experi-ıents.

Of the *à priori* considerations and analogies which have been adduced, will consider only the contention that living vaccines ought to be referred to vaccines which have been sterilized by heat, on the ground hat inoculations with the former furnish close analogies with actual nfections which are known to confer immunity, whereas there are no uch close analogies between these last and inoculations with vaccines vhich have been subjected to the action of heat. To this I could rejoin hat even supposing the temperature of boiling water, which the objector ıas here in view, to produce in a bacterial culture chemical changes as undamental as those produced by that temperature in egg-albumin, a emperature which just suffices for sterilization might quite well leave he chemical constitution of a bacterial culture for all practical purposes ınaltered, just as there is a temperature, short of the coagulating tempera-ure, which will, when applied to a hen's egg, prevent germination, leav-ing the chemical characters of its albuminous substances substantially ınaltered.

If I refrain from all attempt to meet with analogical reasonings of this kind the many other suggestions which have been put forward in connexion with the constitution of bacterial vaccines, it is because I hold that in scientific controversy the proper procedure is always either to bring proof of a contested proposition, or to make *tabula rasa* by demon-strating that such proof is not forthcoming.

Let me now try to show you that the reasoning which has been held to establish that a vaccine of a particular composition is more effective than a vaccine of somewhat other composition has in every case been inconclusive.

Where comparative experiments are undertaken upon two animals or upon two different sets of animals with different vaccines, and where in the one case a more effective response has been obtained than in the other, all that has been established is that the one vaccine employed in a particular dose x has at the date of the test inoculation conferred a greater measure of protection than another vaccine employed in another dose y. Now it is obvious that unless we have assurance that the doses x and y which have been employed in the comparative experiments represent the optimum doses of these vaccines (or doses which stand in each case in the same relation to those optimum doses) we can have no assurance that the survival or non-survival of the test animals is determined by the difference in the vaccine and not by a difference in the dose.

matter of the avoidance of risk. It is by no means certain that the survival of the animals which are treated with serum supplies a correct measure of the immunity of the vaccinated organism which furnishes the serum. It is not even clear that the survival of the vicariously inoculated animals is always, as the theory of this test method would require, the result of "passive," as distinguished from "active immunity." Finally, it is from the practical point of view a not unimportant consideration that the following out of a reaction of immunity by such a method as this might involve the expenditure of a whole hecatomb of animals.

(3) *Consideration of the direct Toxic Effects produced by the Inoculation.*

The idea has widely prevailed in connexion with prophylactic inoculation that the toxic effects which are due directly to the action of the vaccine may be used to gauge the immunising effect. In connexion with this suggestion the following may probably be laid down with confidence.

(*a*) We have in the clinical symptoms only a measure of the intoxication effect which is produced by the vaccine.

(*b*) Immunising response is by no means necessarily proportional to intoxication.

(*c*) There is not yet available any body of experimental data such as would enable us in connexion with any immunisation procedure to expect from a particular quantum of constitutional disturbance a defined amount of immunisation.

It follows that we cannot, in connexion with prophylactic immunisations, accept the clinical symptoms which supervene upon inoculation as a measure, even as an indirect measure, of the immunisation which will be achieved.

(4) *Consideration of the Improvement or Aggravation of the Clinical Condition which follows upon Inoculation.*

The question as to whether, in connexion with therapeutic immunisation, the clinical event may serve to gauge the immunising response which is elicited is a question which cannot profitably be approached until we have made a deeper study of the facts. I will reserve what I have to say on the subject for another occasion,[2] premising here only this, that while it is an axiom that there must, wherever we are dealing with any immunisation process which is therapeutically effective, of necessity be a correlation between immunising response and clinical improvement, the clinical event cannot be recognized as an unfallacious measure of the immunising response, unless we are entitled to assume that the only factor which comes into consideration in connexion with changes in the patient's clinical condition is the occurrence of an immunising response.

[1] Cf. here the discussion of the possibility of active immunisation resulting from the exhibition of so-called "*antibacterial sera*" (*supra*, pp. 309 *et seq.*).
[2] See pp. 436–448, *infra*.

Now this would be a quite unjustified assumption. In the clinical sult we have only the terminal event in a long and intricate chain of usation, and whether that result is amelioration or aggravation, or covery or death, it is in each case the resultant of many factors of which e achievement of a satisfactory immunising response is only one. I need rhaps hardly point out that it would profit a patient nothing to have sponded to an inoculation by an elaboration of antibacterial elements, from any cause the antibacterial agencies which were at disposal in s blood failed to obtain access to the invading bacteria. Nor would it ofit the patient anything in his struggle with disease if his machinery immunisation responded to every call while some other essential part his physiological machinery gave out.

) *Evaluation of the Antibacterial Power of a Sample of the Patient's Blood, and Discussion of the Question as to whether we should aim at a "Complete" or a "Partial Evaluation."*

It having become clear that clinical failures may occur independently any default in the matter of immunising response and that the clinical ent does not in any case furnish a direct measure of the effect of an oculation, we shall be well advised to look elsewhere for that satisctory measure of immunising response which we require. Seeing at all the changes of which we know in connexion with immunisation re changes in the antibacterial power of the blood, we may approriately inquire whether it is not possible directly to measure the effect an inoculation by evaluating the antibacterial power of the blood.

We may perhaps succeed in threading our way through the many itfalls which here lie in our path if we make a point of inquiring whenever ny evaluation of antibacterial power comes up for consideration (*a*) *hether the evaluation in question aims at being a "complete evaluation" r a "partial evaluation,"* (*b*) *what practical value is claimed for the valuation, and* (*c*) *on what grounds that claim is based.* The following onsiderations may perhaps be of assistance in the resolution of these uestions.

Where an immunisator sets out to make a *complete evaluation of he antibacterial power of a sample of blood* he is setting out to obtain measurement which shall be an absolute measure of the resistance of he patient to infection, and which shall be accepted as such, immediately, nd by everybody.

Where immunisator sets out to make a *confessedly partial evaluation f the antibacterial power of a sample of blood* he sets out to obtain a neasurement which—though the expert may value it—will not for he world in general possess any practical significance until it has een established that there is a definite correlation between the condiion of the blood as revealed by such a measurement and the clinical ondition of the patient, and in particular until it has been established hat an increase or diminution of the bacteriotropic element which

is being measured is associated respectively with an aggravation an improvement in the clinical conditions.

When we survey these disadvantages which are incident to a part evaluation of the antibacterial power of a sample of blood, it would first sight seem unquestionable that an immunisator would be well advis to aim in every case at a complete as distinguished from a partial eva ation.

Let me, however, try to show you that we cannot have any assuran that a complete evaluation is in any case practicable; nor, supposi that it were shown to be practicable, could we have any assurance th such a measurement, made as it is *in vitro*, would furnish us with accurate measure of the patient's resistance to infection.

Let us first consider whether anything in the nature of a comple evaluation of the antibacterial power of a sample of blood is in reali practicable.

For a complete evaluation of the antibacterial power of a sample blood there are two prerequisites.

The *first* of these is that our knowledge of the antibacteri agencies of the blood should be exhaustive; that we should know wi precision the rôle which each such agency plays in the destruction bacteria; and that we should have discovered a common denominat such as is required where the sum of the values of a number of diver antibacterial agents has to be arrived at.

The *second* prerequisite is that we should have at command a sati factory technique for measuring the effect of all these antibacterial agencie severally or in combination.

Without going so far as to say that all difficulties in connexio with the elaboration of an adequate technique have been overcome, may be confidently affirmed that what stands between us and a con plete evaluation of the antibacterial power of a sample of blood is muc more fundamental than any question of technique.

When we reflect that it is only very recently that the rôle of the leuco cytes as antibacterial agents has won universal recognition, and whe we look back and remember that the list of bacteriotropic element of the blood might have seemed already complete at a time whe neither the agglutinins nor the opsonins had been discovered, would be clear that it would to-day be venturesome to assert— especially in view of the fact that Wells has shown the blood of infants[1] i sometimes almost devoid of opsonic power—that there are now no furthe antibacterial elements in the blood awaiting discovery.

But even if that were assured it is clear that our knowledge of th antibacterial agencies with which we are acquainted is in many respect incomplete.

We have no assurance that agglutinins actively co-operate in th destruction of bacteria. It is only of the nature of a surmise that they

[1] *Practitioner*, May, 1908.

ay an important rôle in that destruction by the part which they play . promoting phagocytosis. Again, in connexion with the tubercle acillus, and I may add the staphylococcus, we have not as yet any ex- erimental evidence to show whether the opsonins which promote the hagocytosis of these microbes play any active part in their destruction.

We do not even, as a matter of experimental knowledge, know that ese microbes are killed in the interior of the leucocytes.

Above all, where two or more diverse antibacterial agencies come into peration upon a microbe, such as the typhoid bacillus, we cannot say ow much of the antibacterial effect exerted is due to each of the several gencies—how much to phagocytosis and how much to the bacteriolytic ctivity of the blood fluids.

These considerations will, I think, have made it clear to you that 1 the present state of knowledge we *cannot make a complete evaluation f the antibacterial power of a sample of blood, or cannot, at any rate, e sure that it lies in our power to make it.*

I want, further, to try to show you that such *complete evaluation, if prac- icable, would not furnish any absolute measure of a patient's resistance to acterial infection.* Let me, with a view to bringing this home to you, elect for consideration a simple case. As simple a case as any is he case of resistance to staphylococcus infection, for we have here , case where immunity may, for aught we know to the contrary, de- end upon phagocytosis alone. Assuming this to be so, and assuming igures to be available, representing the number of staphylococci hagocytosed by a measured sample of normal blood and again by a neasured sample of a patient's blood, the question would immediately rise as to whether we had here a measure of the phagocytic capacity of the patient in terms of that of the healthy man.

In connexion with this problem I would invite your attention, first, o the fact that in connexion with phagocytic evaluations like those ere in question, there will of necessity have been left out of account eventual differences in the emigrating power of the leucocytes.

Now such differences, if they exist, would in a case of an actual bacterial infection be quite capable of influencing the issue as between the invading microbe and the resisting organism.

Again I would ask you to reflect that where we estimate *in vitro* the relative phagocytic capacities of two bloods, we bring into operation in each case only an infinitesimal fraction of the total force of phagocytes, and only an infinitesimal fraction of the total volume of antibacterial fluids which are in each case available in the organism; and we conduct the experiments under conditions which do not admit either of a rein- forcement of the force of phagocytes which is engaged, or of a supplemen- tation of the antibacterial fluids which are brought into operation.

Now the reinforcement of the phagocytes engaged by new and un- poisoned relays of phagocytes, and the continuous replacement of exhausted antibacterial fluids by unexhausted fluids may quite well

suffice to level up the conditions of an individual who has a less effecti blood to the conditions of an individual who has a more effective bloo

The more we reflect upon the possibilities to which attention has he been drawn, and upon the fact that there must be endless other differenc which escape us, between the conditions which obtain in the case of phagocytic experiment conducted *in vitro* and those which obtain the body in actual infection, the more clearly will it be brought home us—and this is a truth for which Metchnikoff has always contended- that no matter how closely we think we may have reproduced *in vit* the conditions which obtain *in vivo* we can never be sure that w have reproduced all the essential conditions, and it will never be legit mate to treat the measurements obtained *in vitro* as if they held tru without qualification in the living body.

If I have succeeded in bringing home to you that grave, perhap insuperable, difficulties stand in the way of a complete evaluatio of the antibacterial power of the blood, and if I have shown you tha the advantages which such a complete evaluation promises may not b realizable, you will be prepared to consider very seriously the questio as to whether it would not be well in connexion with immunisation pro cedures to abandon all idea of gauging the results of an inoculatio by a complete evaluation of the antibacterial power of the blood an to resort instead to a methodical exploitation of any accurate and non fallacious[1] method of partial evaluation which may prove to give result which are definitely correlated with significant clinical events.

I would venture to point out that I have in all my immunisation work both in prophylactic inoculation against typhoid fever, and in the thera peutic inoculations with which we are here specially concerned, kept thi principle steadily in view. *Abandoning as futile all attempt at a complet evaluation of the antibacterial power of the blood, I have in each case guided myself by measurements of the content of the blood in some one selected bacteriotropic element.*

In the case of my earliest *antityphoid inoculations* [2]—anticipating that there would assuredly prove to be a correlation between the development of high agglutinating power and recovery from, and immunity against, typhoid fever—I guided myself by measurements of the content of the blood in agglutinins, conducted with a technique specially devised for this purpose.

[1] It will be plain that we cannot expect to find evidence of any correlation between the data of a blood test and clinical events unless we are employing an accurate and unfallacious method of arriving at the content of the blood in the particular bacteriotropic element which we have selected for measurement.

Let me observe in passing that in the case of the estimation of the opsonic power the more important fallacies we have to be upon our guard against are: (*a*) the clumping of bacteria under the influence of the agglutinins, (*b*) the dissolution of bacteria under the influence of bacteriolysins, (*c*) the clumping of the corpuscles under the influence of haemaglutinins, (*d*) spontaneous phagocytosis, and (*e*) the intra-cellular digestion of the microbes.

[2] *Lancet*, September 19, 1896; *British Medical Journal*, January 16, 1897.

Anticipating a similar correlation between the development of high bactericidal power and immunisation—I elaborated a technique for the measurement of the bactericidal power of the blood [1] and guided myself then by this more sensitive method [2] in prophylactic inoculation against typhoid fever.

In my first *antistaphylococcus inoculations*—after having elicited that the blood fluids exert no bactericidal effect upon this microbe—and anticipating, therefore, that there would turn out to be a correlation between phagocytic response and clinical improvement—I guided myself [3] by the method of measuring the phagocytic power of the blood which was devised by my friend and fellow-worker, Leishman.

Later, when Douglas and I had elicited the rôle of the blood fluids in connexion with phagocytosis, and the fact that increased phagocytic response occurs in the immunised patient quite independently of any change in the leucocytes, I guided myself in my staphylococcus inoculations by estimations of the opsonic index.[4] Here again I anticipated that there would prove to be a correlation between an increase of the opsonic index and clinical improvement.

Conforming always to the same principle, I guided myself in my earliest *antitubercle inoculations*—following in connexion with these the lead of Koch—by estimations of the tuberculo-agglutinating power.[5] And, as soon as the difficulties which had stood in the way of the accurate determination of the tuberculo-opsonic index had been overcome, I guided myself by this as a more sensitive test.[6]

Exactly the same course was followed in connexion with *prophylactic and therapeutic immunisation against the micrococcus Melitensis.*[7]

Recently in the case of immunisation against most *other microbes* I have gone directly to the estimation of the opsonic index.

You will now realize that the principle upon which as a foundation vaccine-therapy has been built up, is the principle that we should guide ourselves by evaluating in an accurate manner some one selected bacteriotropic element of which we have grounds to believe that its increase in the blood will prove to be correlated with clinical improvement.

Grounds for taking as our Routine guide in connexion with Immunisation Procedures the Estimation of the Opsonic Index.

While, as you now appreciate, vaccine-therapy has been developed on the basis of a partial evaluation of the antibacterial power of the blood, it is not by any means necessarily and indissolubly bound up with the

[1] *Proc. Roy. Soc.*, vol. lxxi, 1902.
[2] *Lancet*, September 14, 1901.
[3] *Lancet*, March 29, 1902; *vide* pp. 199–226.
[4] *Proc. Roy. Soc.*, vol. lxxiv; *vide* pp. 100–111.
[5] *Proc. Roy. Soc.*, vol. lxxiv; *vide* pp. 124–125.
[6] *Proc. Roy. Soc.*, vol. lxxiv, *vide* pp. 126–132.
[7] *Vide* pp. 393 and 395.

estimation of the opsonic index. None the less there are very goo grounds for taking that measurement as our routine guide in connexio with most immunisation procedures.

What is of moment here is, *first*, the fact that the changes in th opsonic index occur, so far as is known, in connexion with immunisatio against all bacteria without distinction, *secondly*, the fact that thes furnish a very sensitive record of immunising response, *thirdly*, the fac that an accurate technique is available for measuring these change and *fourthly*, the fact that there is an overwhelming body of evidenc showing that there is a correlation between the rise and fall of the opsoni index and favourable or, as the case may be, unfavourable changes in th clinical condition of the patient.

As compared with the measurements of the phagocytic power of th blood which can be obtained by the modification of Leishman's metho which was described and made use of by Douglas and myself in ou earlier work,[1] measurements of the opsonic power have an advantage in asmuch as they are not affected by variations in the number of leucocytes or by any variations in the phagocytic activity of these leucocytes, o by such differential variations in the population of leucocytes as woul be produced by the disappearance of older and the advent of younge generations. In the case of opsonic estimations we avoid all suc fallacies; for we concentrate our attention upon a particular change i the blood which stands, as I shall show you, in relation to immunisin response, and we deliberately exclude from consideration all other change which may occur in the blood.

IV.—Detailed Study of the Curves of Immunisation which are Obtained by the Inoculation of Bacterial Vaccines.

It is almost unnecessary for me to describe to you here—for I have already over and over again described them—the outstanding features of the reaction of immunisation which is called forth when a quantum of a bacterial vaccine such as just suffices to produce a sensible constitutional disturbance is administered.

You know that there follows upon such inoculation a fall in the antibacterial power of the blood, and you know that this is succeeded by a rise in the antibacterial power of the blood.

You further know that after a varying interval the antibacterial power of the blood comes down to a level only a little higher than the level of origin and may remain there for a very long period (this is what may be expected in the case of prophylactic inoculations), or it may quickly return to the point where it stood before inoculation (this is what generally happens in the case of an infected organism).

[1] *See p.* 76—*Procedure No.* 1, *and p.* 102—*last paragraph. This method has since been rediscovered and claimed by R. M. Veitch, Journal of Pathology and Bacteriology, Jan.*, 1908; *Hugo Kämmerer, Münch. Med. Wochenschrift*, 1908, *No.* 28; *O. B. Brown, Journal Amer. Med. Assoc., Feb.* 22, 1908, *and others.*

We have here the four events which I have designated as "the ebb" he *negative phase*), "the flow" (*positive phase*), "the backflow" (sub-quent decline of the curve), and the "sustained high tide of immunity" een in connexion with prophylactic inoculations and in connexion with ierapeutic inoculations where recovery is nearly complete.)

This phrase, as already indicated, delineates only the larger features of ie curve, which are revealed by a daily examination of the blood. Where iore frequent and in particular where earlier blood examinations are made wo other features attract attention. The *first* is a transient *initial ise* preceding the negative phase. We may think of this as of an incom-ig wavelet which arrives before the ebb which precedes the main wave f immunity. This wavelet and its position in the general scheme of scillation is shown on the record below (Fig. 1). The *second* feature ; a second sinking away of the curve which occurs subsequent to he positive phase. We may speak of this as the *secondary ebb*.

It is not to be assumed that the form of curve, as set forth in the hart here in question, is universally conformed to.

Where a dose of vaccine which is only just sufficient to produce an ffect on the blood is administered the negative phase is elided, and

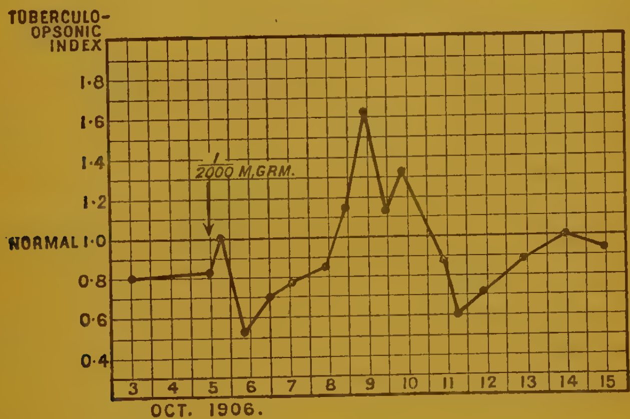


FIG. 1.—Chart showing the changes in the opsonic power of the blood which followed upon the inoculation of $\frac{1}{10000}$[1] milligramme of T.R. into a patient suffering from tuberculous cystitis.

there is registered only a positive phase. The curve in such a case neither rises so high nor does it maintain itself so long above the base line, as in the case where a large quantum of vaccine has been administered.

Where an excessive dose of vaccine is administered—meaning here by an excessive dose a dose whose inoculation produces severe constitu-

[1] For an explanation of the difference between this quantum and that set down on the chart *vide* p. 123, note 2.

tional symptoms—the negative phase is proportionately accentuated an prolonged. Where the quantum of vaccine is immoderately large, th antibacterial power of the blood may be reduced for a period of week The advent of a positive phase may often in such a case be awaited i vain.[1]

Chronology of the different phases of the curve of immunisation.— Having now learned the features of the curves which are obtained afte the inoculation of different quanta of bacterial vaccines, we may furthe study the chronology of the successive incidents.

The traditional view—a view which would seem to have been derive from experience with vaccinia—is that a period of ten days is alway required for the establishment of active immunity. Largely owing t the fact that this view was adopted by Pasteur, it became part of th current religion of bacteriologists, a fixed period of ten days bein always allowed to elapse before animals and men were subjected to tes inoculations or to reinoculation.

So far as I know Haffkine[2] was the first to claim that a condition o immunity was achieved already within 24 hours after the inoculatio of a vaccine. He put forward this claim in connexion with his plagu vaccine, basing his contention on the statistical results of antiplagu inoculations undertaken in India—respectively in the Byculla Gaol i Bombay and in the village of Undhera.

Some years afterwards, the matter having in the meantime bee advanced a step further by the publication, under Haffkine's auspices of evidence pointing to the successful inoculation of patients who wer already in the incubation period of plague—I obtained, in the course o a study of the changes effected by antityphoid inoculation in the bacteri- cidal power of the blood, evidence of development of increased bactericida power in the blood on the day subsequent to the inoculation of moderat doses of antityphoid vaccine.

Following upon this, I obtained, in connexion with my first therapeuti inoculations of staphylococcus vaccine, evidence of increased phagocyti response on the day subsequent to inoculation.

[1] It may be interesting in this connexion to note the following points :—

1. Immoderate negative phases due to over-dosage rarely come under observa- tion except in the case of patients who are new to inoculations. After a prolonged series of inoculations it is the rule (but by no means an invariable rule) to find that a large surplus of vaccine over that which is required to raise the bacteriotropic pressure of the blood can be tolerated.

2. Where by inadvertence an excessive dose of vaccine has been administered it is unnecessary indefinitely to await the return of the bacteriotropic pressure to the normal. In such a case the desired rise can practically always be obtained by reinoculating, as soon as all constitutional symptoms have disappeared, with a minimal dose of vaccine.

3. The initial rise which has been referred to in the text may come into observa- tion also in connexion with the inoculation of an excessive dose of vaccine. It may be provisionally assumed that such initial rise is the response of the organism to that fraction of the total dose which is immediately absorbed.

[2] *Vide* Report of the Indian Plague Commission.

One would have thought that the achievement of an immunising effect within so short a period as 24 hours would have constituted a record.

We have, however, now obtained, in very numerous instances, conclusive evidence of an augmentation of the opsonic power of the blood within an hour after the inoculation of tubercle vaccine, and also trustworthy evidence of associated clinical improvement within that time both in connexion with the inoculation of tubercle vaccine in an infection of the eye, and also in connexion with the treatment of furunculosis by inoculations of staphylococcus vaccine.

The augmentation of the opsonic power and the clinical improvement which are here in question were, as you will appreciate, correlated, in the case where very small doses of vaccine were employed, with the development of the ordinary positive phase, and, in the case where moderate doses of vaccine were employed, with the development of that feature in the curve which was referred to above under the designation of the initial rise.

Question as to whether it would not be possible by Piling one Inoculation upon another to Achieve a Larger Output of Antibacterial Substances and in Association with this a Positive Phase of Longer Duration.

It will be every man's—at any rate, every beginner's—thought that it *ought* to be practicable to achieve a larger output of antibacterial substances, if not by the employment of larger doses of vaccine, then assuredly by piling up inoculation upon inoculation. I have already discussed this question elsewhere,[1] and have pointed out that, while it may be possible in connexion with certain vaccines and the employment of submaximal doses—using the term submaximal doses here to denote doses smaller than those which would give maximal immunising response—to obtain cumulation in the direction of the positive phase, such a result can with other vaccines—and in particular with tubercle vaccines—very rarely, if ever, be obtained.[2]

Where this situation confronts us the proper policy would appear to be to treat each inoculation as an independent event, following up one inoculation by another as soon as the effect of the antecedent one is passing off.

Question as to whether it is Expedient to Employ in Successive Inoculations Progressively Increasing Doses of Vaccine, and Suggestions with Respect to the Regulation of the Dosage.

Close kin to those primitive ideas which suggest that the immunising response *must* increase proportionately with the dose of vaccine employed, and that, failing this, it *must* be possible to obtain an increased immunis-

[1] *Clinical Journal,* May 16, 1906 (*vide* pp. 305–306).

[2] *Transactions of the Royal Medical and Chirurgical Society,* vol. lxxxix (*vide* pp. 272–273).

ing response by piling one inoculation upon the other, is the idea that *must* be possible to achieve a greater yield of protective substances employing in successive inoculations doses of vaccine increasing geometrical progression.

In point of fact, experiment shows clearly, not alone that no adva tage is reaped from such progressive augmentation of the doses of vaccin but that such a system of dosage, if persisted in, culminates in disaste And indeed it is obvious, on *à priori* considerations alone, that whe the dose of vaccine is progressively augmented a point must sooner later be arrived at when the power of immunising response will gi out.

I would therefore put it to you that the proper principle of dosa in connexion with any series of inoculations is never to advance to a larg dose until it has been ascertained that the dose which is being employ is too small to evoke an adequate immunising response.

In actual practice I regulate the dosage of bacterial vaccines f therapeutic purposes in accordance with the following scheme:

Where an examination of the patient's blood taken 24 hours aft inoculation shows that the index has been considerably reduced, I tal it that the smaller dose would have been appropriate.

Where examination of the blood 24 hours after inoculation shows th the index has been raised, and where after the expiration of an interv of a week or ten days the index has fallen back to what it was befo inoculation—the patient having experienced throughout nothing in th form of constitutional disturbance—I take it that a larger dose migl appropriately have been administered.

Where, in association with a slight initial fall after inoculation, tl index is, after the expiration of a week or ten days, found to stand high than it was at the outset, I take it that an appropriate dose has bee administered.

Question as to the Site which Ought to be Selected for the Inoculatio of a Bacterial Vaccine.

A whole array of observations point to the local production of bacteri tropic and vaccinotropic substances generally at the site of inocul tion.

We have, in the first place, the observation that an infinitely great yield of antitoxins—and it would seem also of antibacterial substanc generally—is achieved in horses by the subcutaneous as contrasted wit the intravascular method of inoculation.

We have, further, the observation, in connexion with antityphoi inoculation, that seemingly a more effective immunisation reaction induced in patients who show considerable local reaction at the seat c inoculation than in those who suffer from severe constitutional sympton apart from any appreciable local reaction.

We have further observations to the effect that local immunity ma

be acquired and retained apart from the acquirement or retention of general immunity.

If we may build—as it would seem that we may—upon the aggregate of these observations, it would logically follow that the site of every inoculation which is undertaken for therapeutic purposes deserves to be very carefully considered in connexion with the site of the focus of infection which is to be influenced.

We may speculate as follows. Where protective substances pass into the blood through a channel which does not lead through the focus of infection—let us, for the purpose of fixing our ideas, suppose that focus to be situated in a lymphatic gland—the newly elaborated antibacterial substances will come into operation upon that focus only after they have been diluted by the whole volume of the blood. Where, on the contrary, the inoculation has been made—if I may so express it—*up stream* from the focus of infection—i.e., in some part of the lymph watershed which drains through the focus of infection—the antibacterial substances which are produced at the site of inoculation may, on their passage into the blood, be expected to come into application upon the focus of infection in a concentrated condition.

Pending the time when the place of origin of the bacteriotropic substances which appear in the blood after inoculation shall have been definitely set at rest by some *experimentum crucis*,[1] I have made some tentative therapeutic experiments on the relative efficiency of inoculations undertaken up stream from the focus of infection as compared with inoculations made in regions which were not so related to that focus. I feel satisfied that these experiments—while suggesting that one and the same portion of tissue tires out on too frequent inoculation—have furnished results conformable to those which might have been expected to follow from the theory of the production of bacteriotropic substances at the site of inoculation.

To take only one case out of many, I was much impressed by the fact that the theory of the local production of protective substances suggested a procedure which successfully arrested the spread of a tuberculous affection which long defied treatment by the ordinary method of inoculation. This successful result was obtained in the case of an originally ulcerous process which, while it had completely healed in the centre, was, under the influence of inoculation, spreading at the borders in the form of a ring of indurated tissue. Its arrest and definite cure were achieved when—executing a strategic move—I inoculated the tubercle vaccine in a number of different points disposed circle-wise around the extending ring.

[1] I would throw out in this connexion the suggestion that the production of antibacterial substances at the site of inoculation would be definitely set at rest if, after the inoculation of a bacterial vaccine into a limb, it were shown that an increase of antibacterial substances in the blood could be achieved by either massage or massage combined with the application of a Bier's bandage.

V.—In addition to mastering the Physiology of the Immunising Respons the Immunisator must concern himself also with the following.

Up to this point we have concerned ourselves only with the study o the physiology of immunisation, i.e., with the study of the reaction o the organism to the immunising stimuli and of the methods of applyin those immunising stimuli in such a manner as to achieve and maintain high bacteriotropic pressure in the circulating blood.

We must go further if we are to be in a position to cope successfull with any obstinate bacterial infection.

1. *We must inquire whether, apart from the inoculation of vaccine important changes occur in the bacteriotropic power of the circulating blood and, if so, under what conditions such changes occur.*[1]

2. *We must inquire into the conditions which obtain in the foci in whic bacteria maintain themselves in the organism ; and must ascertain whethe these conditions differ from those in the circulating blood ; and, if they differ in what respect they differ.*

3. *We must, after carefully surveying the whole facts, think out fo ourselves the broad principles which ought to be followed where we set our selves to combat bacterial infections by the agency of the protective element which are furnished by the organism.*

4. *Taking it, as a first therapeutic axiom, that the bacteriotropic pressur of the circulating blood ought in each case to be brought up to, and maintaine at, a high level, we have further to consider, on the one hand, whether thi ought to be effected by the agency of auto-inoculations, or of inoculation with bacterial vaccines ; and, on the other hand, whether inoculations with bacterial vaccines may be employed in cases where we are confronted with "spontaneous auto-inoculations."*

5. *Taking it, as a further therapeutic axiom, that the leucocytes and bacteriotropic substances which are the instruments of immunisation ough to be brought into operation upon the bacteria in the focus of infection, we must consider by what means this may in each case be effected.*

Having formulated our programme, we may take up the study of these subject-matters *seriatim*. We may begin by a study of auto-inoculations.

(1)—Auto-inoculations.

It will be obvious to consideration that intoxication phenomena and immunising responses exactly similar to those which supervene upon the inoculation of bacterial vaccine, must in the ordinary course occur whenever bacterial products, or as the case may be bacteria, escape from localized foci of bacterial infection and pass into the circulation.

It will be clear that it must be by the agency of such auto-inoculations that the machinery of immunisation is set in motion in bacterial infections,

[1] The doctrine of auto-inoculations is developed in the next subsection.

and that it must be by the agency of immunising responses to such auto-inoculations that spontaneous cures are achieved.

If you will now bring this into relation with the facts to which I drew your attention when I was discussing the "expectant treatment" of bacterial disease, you will appreciate that generalized infections must be characterized by frequent, perhaps continuous, auto-inoculations; that strictly localized infections must be characterized by an absence of auto-inoculations; and that there must be an intermediate class of localized infections—infections which are associated with occasional constitutional disturbance—where there must be occasional auto-inoculations.

And again you will see that we have here an explanation of the fact, that something is to be hoped for from expectant treatment in generalized infections while little or nothing is to be hoped from it in connexion with strictly localized infections.

And it will now have become clear to you that the something unknown which the "medical attendant" sits down to "expect" in bacterial infections is an effective auto-inoculation—or rather such a series of properly graduated and properly timed auto-inoculations as shall evoke in the organism of the patient adequate immunising responses.

Up to the present we have thought only of *spontaneous auto-inoculations*. We have to consider now another class of auto-inoculations, to wit, artificially *induced auto-inoculations*.

Taking our departure from an illuminating observation made by my collaborator, Dr. J. Freeman, in connexion with the effects produced on the blood by the massage of a gonococcal joint, we have at St. Mary's Hospital during the last twelvemonth devoted ourselves to a systematic study of the conditions under which auto-inoculations can be produced in persons affected with localized bacterial infections.

We have been able to show that auto-inoculations follow upon all active and passive movements which affect a focus of infection, and upon all vascular changes which activate the lymph stream in such a focus.

Evidence has been obtained of the production of auto-inoculations by massage and extirpation operations affecting tuberculous glands; by passive extension, massage, and divers surgical operations affecting tuberculous and gonococcal joints; and by scraping operations undertaken in connexion with tuberculous caries and staphyloccocal osteomyelitis.

Again, evidence has been obtained of the production of artificial auto-inoculations in phthisical patients when they were called upon to breathe deeply and when they were examined by percussion and auscultation.

We have also in connexion with a laryngeal affection seen an auto-inoculation supervene upon reading aloud.

Again, we have obtained evidence that auto-inoculation may follow upon walking exercise in the case of patients affected with tuberculous disease of the lungs or of the bones or joints of the lower extremity or with severe tuberculous epididymitis.

We have also obtained evidence that in the case of patients with spina caries auto-inoculations are induced by a change from the recumbent to the sitting posture and from the sitting to the erect posture.

Lastly, we have evidence that auto-inoculations are produced both by active and passive hyperæmia (application of hot fomentations and of Bier's bandages) to limbs affected with tubercle and streptococcus respectively.

Assistance which can be Obtained from Purposively Induced Auto-inoculations in the Diagnosis of Obscure Cases of Localized Bacterial Infection.

Let me for a moment here digress from my main theme to consider the assistance which may be derived from the induction of artificial auto-inoculation where a problem of diagnosis confronts us.

Rightly understood, all methods of bacteriological diagnosis in which we arrive at the nature of the infection by a process of induction from a measurement of the bacteriotropic content of the blood are methods which have as their aim the detection of the changes which are produced in the blood by the agency of auto-inoculations.

The following three examples will suffice to make this clear.

The agglutination test in typhoid fever—credit for which was claimed by Widal on the plea that the increased agglutinating content of the serum was something quite unrelated to an immunising response evoked by an auto-inoculation [1]—is now by common consent recognized to be a test which depends—as Gruber, with his collaborators Durham and Grünbaum, from the very first discerned—on the detection of products of immunity produced in response to an auto-inoculation.

The "test for thermostable opsonins," whose diagnostic value was first demonstrated by Reid and myself, furnishes in like manner evidence of an immunising response to a foregoing auto-inoculation or, as the case may be, to an inoculation of the corresponding bacterial vaccine.

Exactly the same thing holds true of Bordet's "absorption of complement test" recently so ingeniously exploited by Wassermann.

It is clear that in all these cases diagnosis has perforce to wait upon spontaneous auto-inoculation, and that perforce it has to wait also upon immunising response. Now both the one and the other of these may, as we have seen, in connexion with localized infections, be indefinitely deferred.

It accordingly marks a distinct step in advance when we come to realize that we can, in the case where the focus of infection is accessible, supply the place of the spontaneous auto-inoculation which makes default by the induction of an artificial auto-inoculation.

Let me say in this connexion that we have—by the agency of massage,

[1] "Pour arriver à la conception de séro-diagnostic j'ai dû précisément commencer par me débarasser de l'idée erronée que la réaction agglutinante était une réaction d'immunité."—*Widal*, Annales de l'Institut Pasteur, vol. xi, 1897, p. 359.

active muscular movements, Bier's bandaging, or, as the case may be, other methods of artificial auto-inoculation, combined in each case with the measurement of the opsonic index before and after such event, obtained diagnostic results which have up to the present invariably been borne out by the subsequent history of the case.

I would note here—for it is germane to the subject-matter under discussion—that we have had recourse to auto-inoculations associated with measurements of the opsonic index not alone for the preliminary diagnosis of the bacterial infection, but also for the purpose of obtaining information with regard to the progress of the patient.

Where an artificial inoculation can no longer be induced in a focus which previously could be made to influence the blood we are entitled to conclude that the focus of infection is extinct.

Where an auto-inoculation can still be induced we may be assured that the focus of infection is still aglow.

(2)—Conditions which Obtain in the Foci of Bacterial Infection.

I have already often pointed out that the foci in which bacteria cultivate themselves are in every case "*foci of lowered bacteriotropic pressure*," and that the deficit of antibacterial substances in such foci can be accounted for by the fact that bacteriotropic substances are absorbed whenever blood fluids come in contact with bacteria, and by the fact that in the case of foci which are cut off from the blood stream the conveyance of bacteriotropic substances to the focus of infection by the lymph stream can only rarely keep pace with the afore-mentioned absorption. This premised with regard to the conditions which are common to all bacterial foci, I may here conveniently direct attention to *special conditions which obtain* (*a*) *in infections of serous membranes where serous effusion has taken place*, (*b*) *in abscesses*, (*c*) *in sinuses, and* (*d*) *in association with brawny swelling of the subcutaneous tissues.*

(*a*) *Conditions which obtain in the case where bacteria are growing in, or in contact with, serous effusions.*—These are well exhibited in connexion with tuberculous peritonitis. Here, as my fellow-worker, Douglas, and I have already shown, the ascitic fluid has in every case a much lower opsonic index than the circulating blood. It follows that the bacteria which are cultivating themselves in, or in contact with, such ascitic fluid are not exposed to the full bacteriotropic pressure of the circulating blood. We have here, as I have already pointed out, an explanation of the success which has attended tapping, and in particular laparotomy, in connexion with tuberculous peritonitis. That success is satisfactorily accounted for by the replacement of a lymph which has by stagnating in the focus of infection forfeited much of its antibacterial virtue by a fluid of higher efficacy freshly derived from the circulating blood. Manifestly we should be neglecting a very important element in the treatment if, while aiming at the destruction of bacteria

in a serous membrane by processes of immunisation, we wer[e] to fail to take into account the fact that the bacteria which are th[e] object of our attack are cultivating themselves under a lowered bacterio[-] tropic pressure.

(*b*) *Conditions which obtain in abscesses.*—In abscesses the condi[-] tions are more complicated. Here we have to take into account no[t] only the absorption of bacteriotropic substances by bacteria, but als[o] another factor which does not come into consideration in the case o[f] ordinary serous effusions. This factor is the liberation of a tryptic fer[-] ment from the leucocytes. Such liberation occurs, as Opie has shown[,] whenever these formed elements disintegrate in pus. It is clear tha[t] as soon as—under the influence of autolysis or bacterial action—leucocyte[s] have disintegrated in an abscess in numbers sufficient to abolish the opsoni[c] and antitryptic power of the surrounding fluid, not only the norma[l] bacteriotropic defence of the blood fluids, but also the leucocyte defence wil[l] be thrown out of gear. I have neither the time nor the data to discuss with you here certain important but incidental issues which suggest themselves in this connexion, in particular the issue as to whether the liberation of tryptic ferment in the abscess—which Opie has brought into association with the destructive and burrowing action of pus—may not also account for the paralysis of all phagocytic effort which sooner or later overtakes the leucocytes in every focus of suppuration and for the frequent sterilization of the contents of an abscess. It is, however, incumbent on me to point out to you that where we are aiming at the destruction of the bacteria in a suppurating focus by the agency of opsonins and leucocytes, and are aiming at the same time at the safe-guarding of the tissues from the digestive action of the pus, it would be futile to attempt this task without making provision for the replacement of the tryptic and non-opsonic pus fluid by an antitryptic and opsonic fluid freshly derived from the circulating blood.

(*c*) *The conditions which obtain in sinuses.*—The conditions with which we have to deal in a sinus where that sinus is freely discharging pus are, I take it, essentially similar to those which prevail in an abscess which is discharging without emptying itself. In other words, we have a pus fluid which possesses a low opsonic power, and which exerts upon the tissues a digestive effect—a digestive effect which makes itself manifest to the eye in the case of a discharging sinus in the sodden and unhealthy appearance of the skin around the orifice. In the case of a choked sinus we have to deal with conditions which might be compared to those which would obtain in a well if the water which originally flowed into it had deposited an insoluble element in such a manner as to choke up all the conduits of supply. We can easily conceive how, on the walls and floor of such a well, forms of life might maintain themselves which would be quite incompetent to penetrate into the surrounding soil, or to hold their own in the face of a copious flow of water from the soil into the well. A dry sinus is, if I understand the situation aright, analogous

to just such a choked well, the obstacles to the inflow of lymph being, on the one hand, the density of the granulation tissue, and on the other hand, the lining membrane of fibrin which clothes the walls of the sinus. If these are in reality the conditions under which bacteria maintain themselves in a sinus, we shall need for their dislodgment something more than a mere increase of the bacteriotropic power of the blood and circulating lymph.

(*d*) *The conditions which obtain in "brawny swelling*."—Next in order we have to consider the case where we have a focus of bacterial growth in tissues which are affected with "brawny swelling." I take it that in brawny swelling we have conditions which are—in the respect that the bacterial growth is cut off from the blood and lymph stream—analogous to those which have just been under consideration in connexion with sinuses. It is in the nature of a minor difference only that in brawny swelling the bacterial growth is isolated from the blood and lymph stream by the clotting of the lymph in the lymph spaces, while in the case of a dry sinus the isolation of the bacteria is brought about by the clotting of the lymph on the surface of granulation tissue.

VI.—On the two Broad Principles which ought to guide us where we set ourselves to combat Bacterial Infections by the Agency of the Protective Elements which are furnished by the Organism.

The fundamental principles which ought to inform our therapeutic operations where we undertake to combat bacterial infections by weapons taken from the armoury of the organism may conveniently be expressed in two general principles.

These may be formulated as follows:—

Principle 1.—*Therapeutic immunisation should be resorted to in every case where the antibacterial power of a patient's blood falls below the standard which is attained where the organism is making effective response to infection.*

Principle 2.—*Where the blood is rich in antibacterial elements a fuller lymph stream should be determined to the affected part in order that the antibacterial elements and leucocytes of the blood may come into effective operation in the extravascular focus of infection.*

(1) Comparison between the Therapeutic Policy which is embodied in these two Principles, and the Policies which are embodied in the Measures currently employed in the Treatment of Bacterial Infections.

It will, I think, be useful if, in connexion with the therapeutic programme which is embodied in these precepts, you will let me try to show you both wherein it agrees, and wherein it differs from the therapeutic measures which are currently employed in the treatment of bacterial infection.

(*a*) Where "*expectant treatment*" is resorted to, the above programme

is departed from, first, in the respect that the measurements of the anti-bacterial power of the blood which are postulated in *Principle* 1 are omitted, and, secondly, in the respect that the physician contents himself with awaiting spontaneous auto-inoculations and immunising responses, whereas *Principle* 1 demands that he shall, where spontaneous immunisation makes default, address himself to the task of immunisation.

(*b*) In *serum-therapy* an attempt is made to carry out the therapeutic programme of *Principle* 1. That attempt, in the form in which it is usually made in connexion with septicaemic infections is, however, an altogether unintelligent attempt. It is unintelligent, *first*, in the respect that the investigation of the patient's blood is consistently omitted ; and, *secondly* in the respect that the physician makes here four unverified assumptions. He makes, *first*, the assumption that the organism of the animal which supplies the serum will have responded with an immunising reaction to the immoderate quantities of bacterial vaccines which the serum manufacturer is wont to employ ; *secondly*, the assumption that a fractional part of such output of antibacterial substances as has been obtained in the horse will suffice to produce a therapeutic effect in man ; *thirdly*, the assumption that the antibacterial elements which are postulated to be present in the horse serum will exert a therapeutic effect on man in spite of the fact that they are foreign to the human organism ; and *fourthly*, the assumption that these antibacterial elements will exert this therapeutic effect notwithstanding the very high dilution in which the serum comes into application.

Let it be noted that where vaccine-therapy is the form of therapeutic immunisation which is employed, the antibacterial substances which are obtained are native to the patient's organism ; further, that the total yield of these substances can be brought to bear on the infecting microbes in the concentration in which they are available in the blood.

(*c*) In the case where an attempt is made to deal with bacterial infection by the *extirpation of the obtrusive focus or foci of infection* it may happen upon occasion that the programme of *Precept* 1 may without the knowledge of the surgeon be carried out. This will happen when in response to one of those auto-inoculations, which are, as we have seen, incident to surgical operations, an immunising response has been evoked in the organism of the patient.

Let it, however, be noted that even where everything is favourable we cannot hope much from such an isolated and unregulated auto-inoculation as would be associated with a surgical operation.

(*d*) Where the surgeon *cuts down upon pus and drains*, or where he makes, in connexion with a carbuncle or with " brawny infiltration," free incision into tissue spaces which are blocked with inflammatory exudation, supplementing in each case the work of his knife by *hot fomentations* or *cupping*, he is carrying out the programme of *Principle* 2 by provid-

ng opportunity for a free streaming of lymph through the focus of nfection.

Let it be noted that while the carrying out of so much of our therapeutic programme as applies to the determination of lymph to the focus of infection may, where the antibacterial power of the patient's blood is satisfactory, by itself suffice to extinguish the infection, such success will be almost out of question where the antibacterial power of the patient's blood fails to come up to the normal standard.[1] Where this is the case, *Principle* 1, which enjoins resort to the therapeutic immunisation, will be found to be not merely a counsel of perfection but a dictate of absolute obligation.

(*e*) In the case where a *Bier's bandage*, or a hot fomentation, or any other device for activating the lymph stream is applied *in the absence of an external outflow for the lymph* there will be carried out in association with the programme of conveying antibacterial elements into the focus of infection also a programme of auto-inoculation. As between that programme and the programme of therapeutic immunisation which is prescribed in *Principle* 2 there are quite important differences. These differences will be explained when we come to consider the question whether auto-inoculations can be substituted for inoculations with bacterial vaccines.

Let us, however, first consider certain more general points in connexion with our therapeutic programme.

Rationale of the Policy which is embodied in the Two Therapeutic Principles formulated above.

By the *achievement and maintenance of a high bacteriotropic pressure in the circulating blood* the following advantages will be realized :—

(*a*) The citadel of the circulating blood will be held secure against septicæmic invasion.

(*b*) Bacteria when carried into the blood will be killed there instead of being carried from point to point unharmed, and in a condition to establish new foci of infection.[2]

(*c*) There will be at disposal in the blood a reservoir of antibacterial fluid of satisfactory potency which will be available for the purposes of flushing any bacterial nidus in the tissues.

By the *determination of lymph to the focus of infection* the full bacteriotropic pressure of the circulating blood—whatever that may happen to be—

[1] The futility of operating against bacteria with a lymph of low bacteriotropic pressure—a fact which comes home to every one who examines the blood of patients who fail to get well under ordinary treatment—is conclusively established in a paper by Dr. Bulloch dealing with the tuberculo-opsonic indices of 100 lupus patients who were under treatment in the Finsen light department of the London Hospital. It is clearly shown in that paper that the Finsen treatment of lupus fails in cases where tuberculo-opsonic power stands at an abnormally low level (*vide supra*, pp. 146–149).

[2] *Vide, as an illustration of the importance of this point, infra, pp.* 398–399, *and p.* 412.

will, as has already been indicated, be brought into operation upon th[e] bacteria which would otherwise be exposed only to the lower bacteriotropi[c] pressure which obtains in the focus of infection.

Question as to which of these two Therapeutic Principles is to be followe[d] in those Cases where they come into Conflict.

As has already been incidentally suggested at the outset of thi[s] lecture, we have to take into consideration in connexion with the activatio[n] of the lymph stream in the ordinary case—the case where no externa[l] outlet for the lymph is provided—on the one hand, the antibacteria[l] effect which the lymph stream will exert upon the invading bacteria, and on the other hand, the auto-inoculation effect which will be exerted by the dissemination in the organism of the bacterial products which have been washed out of the focus of infection.

Now while such auto-inoculation may carry with it advantage[s] (as may be seen when, in connexion with Bier's treatment, improvement manifests itself in foci anatomically remote from the seat of application of the bandage) it may often happen that the lymph stream which is induced in the focus of infection will convey into the blood an excessive quantum of bacterial products. In such a case the auto-inoculation will result in a lowering of the bacteriotropic pressure of the blood and upon occasion in constitutional disturbance.

Where we have to choose between lowering the bacteriotropic pressure of the circulating blood by an excessive auto-inoculation, and leaving the bacteria in a localized focus of infection for the nonce unmolested, we ought unhesitatingly to elect for the latter alternative. The safeguarding of the citadel of the circulating blood against septicaemic invasion, and the building up of a barrier in the blood against the passage of living bacteria from one point of the system to another, are considerations which must outweigh all others. And be it noted, even if we were prepared to jeopardize the life of the patient for the sake of molesting the bacteria in the focus of infection such policy would carry with it its own Nemesis; for once the quality of the blood fluids in the main reservoir had been tampered with, immediately all power of operating effectually upon the bacteria in the focus of infection would be lost.

(2) Do Artificially-induced Auto-inoculations furnish a better Agency for raising the Bacteriotropic Power of the Blood against the Infecting Microbe than Inoculations of Bacterial Vaccines?

In the following respects auto-inoculations would appear to have the advantage over inoculations of bacterial vaccines.

(*a*) Where we are employing auto-inoculations we must inevitably be employing the correct vaccine, or in the case of a mixed infection the correct mixture of vaccines.

(*b*) Our therapeutic operations are not—when we proceed by the nethod of auto-inoculation—as they are when we proceed by the method of vaccine-therapy—limited by our power of cultivating the infecting nicro-organisms on artificial media.

(*c*) Treatment by auto-inoculation may in every case be begun withut any preliminary diagnostic work, and without the delay which s inevitable where a special vaccine has to be prepared.

(*d*) The draining off from the focus of infection of the lymph which s impregnated with bacterial products, and its replacement by lymph reshly derived from the blood stream, may be expected to exert a benefiial effect upon that focus.

These advantages are, however, more than outweighed by the following isadvantages.

(*a*) In the case of auto-inoculations we operate with living cultures. 'he activated lymph stream may accordingly carry into the blood stream, ıot only bacterial products, but also living bacteria.

(*b*) In the case of auto-inoculations we are operating with unıeasured, and therefore often ill-adjusted, doses of bacteria and their roducts.

Where we have to deal with a very considerable focus of infection, r, failing this, where in connexion with a smaller focus the irrigation vith lymph is very searching and unduly prolonged, there will be washed nto the general blood and lymph stream an excessive dose of the bacterial roducts.

Where, on the other hand, we have to deal with a small focus of inection, or where in the case of a larger focus irrigation with lymph is ontinued for too short a time, too small a vaccinating dose will come into ıpplication.

Again, where by reason of a gradual restriction of bacterial growth ffected by immunisation, or where by reason of a repeated draining off f bacterial products under the influence of Bier's treatment or of massage, liminishing quantities of bacterial products are available in the focus of nfection, there will come into application diminishing doses of vaccinating lements, while there may quite well be required, for the maintenance f adequate immunising responses, undiminished, or increasing, doses f these elements.

(*c*) Auto-inoculations are not everywhere practicable. Active and assive hyperæmia can be conveniently applied only in the case of foci vhich are positioned in the extremities or in other accessible situations; vhile active and passive movements are applicable as auto-inoculatng agents only where such movements do not involve pain or local njury.

(*d*) As compared with immunisation by bacterial vaccines hypodermiclly inoculated, immunisation by auto-inoculations is, it would seem, lways more expensive to the patient—expensive in the sense that the

patient obtains for one and the same equivalent of intoxication a smalle yield of bacteriotropic substances.

(*e*) Finally: The demands which are made upon the patient's tim and the work which is thrown upon the physician are, in the case whe auto-inoculation methods are employed under the control of bloo examinations, much more serious than in the case where the patient inoculated with bacterial vaccines.

Can Inoculations with Bacterial Vaccines be undertaken in Bacteri Infections which are associated with Spontaneous Auto-inoculations ?

The issue as to whether inoculations of bacterial vaccines can b undertaken in bacterial infections which are associated with auto-inocu lations would at first sight appear to be a perfectly simple issue. I reality we have to consider here a number of different problems. W have to distinguish (*a*), *the case where we have microbes growing in localized focus of infection and where auto-inoculations are superinduce when a lymph stream courses rapidly through that focus, and* (*b*) *the cas where the infecting microbes are cultivating themselves in the circulatin blood, or in direct anatomical relation to this, producing "spontaneous aut inoculations" whenever the bacteriotropic pressure of the blood falls.*

(*a*) *Case of a localized infection with "superinduced auto-inoculations.* Here treatment ought, I take it, to be directed, *first*, to placing a chec upon the auto-inoculations, and, *secondly*, to following up by vaccine therapy, carefully regulated, any advantage which the patient may hav derived from his past auto-inoculations.

The first of these objects may in many cases be achieved by keep ing the patient absolutely at rest, and by avoiding in particular a muscular movements which might send lymph through the focus o infection.

Where rest by itself does not suffice to cut off the auto-inoculation it may prove possible further to diminish the lymph stream by taking ad vantage of the blood-inspissating and anti-lymphagogic action of calciu salts.

The great principle which we have to keep in view in connexion wit the following up of auto-inoculations by inoculations of bacterial vaccine is that in every case, before the employment of bacterial vaccines is under taken, the patient should be permitted to derive all the advantage tha he can from his past auto-inoculations. Where the patient's index i seriously reduced by reason of a foregoing auto-inoculation an interval ough to be allowed to him for recovery before any further inoculation is made And again where the patient's opsonic index is already well above th standard of the normal, inoculations ought to be postponed till th index is on the decline.

(*b*) *Case of a generalized infection with spontaneous auto-inoculations*

In the case of *a septicaemia which is evoking from the organis satisfactory immunising responses*, the proper policy would appear to b

policy of abstention from all interference. In other words here—and, I ay add, here alone—the expectant treatment would appear to be in place.

In the case *of a septicaemia which fails to evoke any, or which evokes nly very unsatisfactory immunising responses* I would suggest that an ttempt should be made to call forth immunising responses by inocula-ions of bacterial vaccines.

To this proposal it might very naturally be objected that, inasmuch s the vaccinating bacterial products are here already circulating in the lood, producing an intoxication, it would be unreasonable to expect rom the incorporation of further bacterial elements anything more han an aggravation of that intoxication.

Let me, however, try to show you that the employment of vaccines these cases is not the unreasonable proceeding that it might at first ight appear.

I would call back to your mind here what was said above on the sub-ect of the probable place of origin of the antibacterial substances.

If I am right in supposing that the bacteriotropic substances are anufactured in the tissues at the seat of inoculation, consideration will how that the conditions for successful immunisation must be less favourable hen the vaccinating elements are thrown into the circulating blood than hen they are inoculated directly into the tissues. In the case where he vaccines are introduced into the circulation they will come into appli-ation upon the tissues only after they have been diluted by the whole olume of the circulating blood; where they are inoculated directly into the issues they will come into application upon these in a concentrated form.

It is accordingly not irrational to assume that there would be a possi-ility of a septicaemic patient deriving in this respect advantage from he inoculation of bacterial vaccines.

There still remains the objection that the inoculation of bacterial accines might aggravate his intoxication.

The rejoinder to this objection is, I think, suggested by the considera-ion that local toxic effects on the tissues—such as are produced by the ubcutaneous inoculation of vaccines—and a local elaboration of bacterio-ropic substances—such as we have reason to believe follows upon inocu-ation—would be inexplicable apart from a holding back of the toxic ubstances in the tissues.

Now if toxic substances are held back in the tissues, it follows that he incorporation of an aliquot quantum of vaccine into the tissues must roduce less intoxication than the inoculation of that same quantum of accine directly into the blood stream.

I would put it to you in view of these considerations that the question s to whether vaccine-therapy can, or cannot, be successfully employed n connexion with septicaemic disease is a question which ought not to be rejudged. It is a question which can be decided only by actual trial.[1]

[1] *Vide here the results of such vaccine-therapy set forth on pp.* 392–409, *infra.*

(3) **Consideration of the Therapeutic Measures which are Appropria where we desire to bring the Antibacterial Agencies of the Blood in Application upon the Bacteria in the Focus of Infection.**

While I must needs postpone to another occasion[1] a full expositic of the therapeutic principles which the immunisator ought to take a his guide where the antibacterial agencies of the blood are to be brougl into application upon microbes which have established themselves in th tissues, I may here briefly summarize the principles as follows :—

We must provide for the conveyance of bacteriotropic substances in the infected region.

In the case where an accumulation of stagnant fluid in the focus infection effectually prevents the entrance of bacteriotropic substances w must as a preliminary measure draw off the fluid which occupies that focu

In the case where there are other mechanical obstacles to the free streamin of lymph through the focus of infection we must remove those obstacles.

(*a*) *Conveyance of bacteriotropic substances to the infected region.*— The douching of the focus of infection by a stream of lymph fresh fro the blood vessels can in the case where the tissues are uninjured and th lymphatic channels are unobstructed, be effected by determining by th agency of heat, or any other rubefacient, a larger blood supply to the regio affected. It can also—as in Bier's method—be effected by banking u the blood in the veins in such a manner as to increase the hydrauli pressure in the capillaries.

(*b*) *Removal of stagnating fluid from the focus of infection in the cas where this prevents the lymph which transudes from the blood vessels findin proper access to the infecting bacteria.* In the case where the lymph ca effectively make its way through the focus of infection, permeating ever part of it, access of the bacteriotropic substances to the infecting bacteri will manifestly be provided for by the activation of the lymph strea quite apart from any operative interference. In such a case the stagnan fluid which occupies the bacterial focus will—whether for good or fo ill—be driven on into the general circulation by the *vis a tergo* exercise upon it by the activated lymph stream. Where this method of dispersio is inapplicable, or where for any reason it is contra-indicated, the evacu ation of the stagnating fluid by operative measures will obviously b desirable. In the case where we have in an abscess a tryptic fluid whic is eating its way into the surrounding tissues[2] such evacuation, will no only be desirable but imperative.

Where we elect to employ the method of evacuation, as distinguishe

[1] *Vide infra, pp.* 452 *et seq.*

[2] It is perhaps worth noting that the fact that an abscess gives fluctuatio does not by any means furnish sufficient warrant for concluding that it contain a tryptic fluid and that operative measures must be resorted to. Again, even wher the contents of an abscess have by the disintegration of leucocytes already acquire tryptic powers, it may still be practicable to abolish that tryptic power and t effect resolution by leading into that abscess cavity a sufficient quantum of antitrypti lymph.

rom the method of dispersion, our choice will lie between simple incision, ıcision followed by cupping—as advocated by Klapp—and evacua- ion by aspiration. This last method has, I submit, an advantage over ll others in the respect that it does away with all scarring and minimises oth the risk of the entrance of bacteria from without, and the risk of uto-infection of the edges of the wound. It also secures, more effectively ıan any method of incision and drainage, what is in the case of an abscess ıe obvious desideratum, to wit, the filling up of the evacuated cavity ith an antitryptic and opsonic lymph which will both inhibit bacterial rowth and arrest further digestive destruction of the tissues.

(c) *Removal of obstacles to the free streaming of lymph through a focus f infection.*—We have seen above that a deficient outflow of lymph and ıe formation of a lining of fibrin on its walls are in the case of a sinus ıvourable to the survival of the infecting microbes. I am accustomed ɔ combat these conditions by introducing into every dry sinus a solution f 0·5 per cent. citrate of soda and 5 per cent. sodium chloride. I ıay explain that the citrate of soda, by decalcifying the lymph, pre- ents coagulation and scabbing; and that the salt, acting by osmosis, ıuses fluid to transude from the blood vessels. Under the influence of ıis application a clear lymph wells out and the local conditions rapidly nprove.

The situation in the case where we have a carbuncle or brawny velling of the subcutaneous tissues being, in the respect that the ʼmph stream is obstructed, essentially similar to those which have ıst been considered, it must be treated on the same principle.

I may perhaps in this connexion make brief reference to a case f Ludwig's angina which came not long ago under my observation. he patient, a middle-aged man, had in the first instance developed what as taken for an indolent furuncle in the parotid region. When, after onsiderable delay, this was incised no trace of pus was met with, and the ssues were found to be everywhere dry and infiltrated. They remained ı this condition, and the wound showed absolutely no disposition to eal.

Two weeks later the patient, who up to that time had been taking ıtdoor exercise, was suddenly taken seriously ill, and the brawny swell- ıg, which up to that time had been limited to a patch on the left cheek, pread rapidly round under the jaw from one ear to another.

A surgeon now carried a series of vertical incisions deep down into ıe indurated tissues. Twenty-four hours afterwards the patient had ıpsed into a condition of low delirium; and the local conditions showed ɔ sign of improvement.

When brought to see him I could not, even at the bottom of the ıping incisions, find sufficient moisture to fill the loop of a platinum eedle. Film preparations obtained by pressing cover-glasses against ıe sides of the wound showed very abundant streptococci, and only ere and there a leucocyte. Blood from a vein at the elbow, drawn

with the intention of making a culture, clotted instantaneously in th syringe.

It was immediately clear that what was most urgently required this case was, not that further means of antibacterial defence should l furnished to the patient, but that such means of antibacterial defen as were already at his disposal should be brought into application upo the streptococci in the focus of infection. With a blood so viscid an coagulable as was that of this patient, it was inconceivable that an lymph should transude into his tissues. It was to be expected, als that any transuded lymph would immediately clot.

Influenced by these considerations, I prescribed 60-grain (4 gramm doses of citric acid[1] every three hours. Six hours after the treatment ha been begun lymph began to ooze into the wounds, and by next mor ing all the wounds had begun to bleed. The administration of citric ac was now suspended.

A culture of the infecting microbes having now been obtained, th opsonic index of the patient's blood was determined. This working ou at 1·8, and very distinct amelioration having taken place in the patient symptoms, I did not think it necessary to resort to inoculation trea ment. Nor did afterwards any occasion for immunising interventio arise, the patient making continuous and rapid progress to comple recovery.

There is, I think, a lesson in this case which we shall do well to tak to heart in connexion with all conditions where the access of lymph t the infected tissues is difficult.

With the discussion of these special therapeutic problems I have a last brought to a close my exposition of the main principles of vaccin therapy so far as these are at present clear to me. You will, perhap expect me to say in conclusion one or two words on the subject of th results which have been achieved by the application of this method i actual practice.

VII.—General Discussion of the Question as to how the Achievements of Vaccine-Therapy ought to be evidenced and adjudicated upon.

Although, as I am well aware, nothing can be more irksome than t be called away to consider abstract principles where one desires to lear of actual achievements, I would venture to urge upon you that it woul be well before adjudicating upon the value of vaccine-therapy to as yourselves what results you may within reason expect from the metho and what proof of its achievements you may legitimately require.

You will perhaps respond in your thoughts that surely a correc judgment on the value of vaccine-therapy might be most easily arrive at, if the case for and the case against this new therapeutic method wer

[1] *Vide Wright and Knapp,* "*Lancet,*" *December* 6, 1902, *p.* 1531; *Medico-Chirurg Transactions, vol.* 86; *and Wright and Paramore,* "*Lancet,*" *October* 14, 1905.

argued before you by able advocates representing the extreme positions of enthusiasm and scepticism. You think very probably, that in the course of such a discussion the legitimacy of every postulate would be investigated, and that by each party every argument would be brought forward which might tell in favour of his own side and against that of his opponent.

I have no doubt that that would be so. But none the less I would deprecate polemical discussion as an agency for preparing the mind for the task of adjudication, and as a means of arriving at scientific truth. I go further. If a man has put forward his case in such a way as to give an opening for polemical attack, I hold that that man is always deserving of blame.

If such a one—so I put the matter to myself—had been careful not to advance any statement without adequate proof, if he had kept his eyes open to every issue which ought to have been considered, if he had explained everything which stood in need of explanation, if he had never lapsed into ambiguous language, and lastly, if he had had sufficient discernment to anticipate the criticisms of the unintelligent and biased, assuredly, he would not then have furnished openings for polemical attack.

I have, too often, had to reproach myself for coming short in every one of these respects. But here I have redoubled my efforts. In connexion with every conclusion which I have arrived at, I have here endeavoured to set forth the chain of reasoning which has led me to that conclusion. I have still to try to reply by anticipation to the censures which I shall draw upon myself by neglecting to furnish to you evidence of the value of vaccine-therapy in accordance with certain required forms, and for addressing myself instead to the task of furnishing you with other and more convincing evidence.

Unreasonable and unconsidered Requirements in connexion with the Evidence to be adduced in proof of the Value of Vaccine-Therapy.

The following are the criticisms which have been made, or may be expected, in connexion with the evidence which has been furnished of the value of vaccine-therapy.

(*a*) *The clinical data with respect to the results of vaccine-therapy ought to be kept quite separate from, and ought to be considered quite without reference to, the data which are furnished by blood examination.*

(*b*) *In connexion with vaccine-therapy nothing short of a complete cure ought in any case to be accepted as evidence of success; again, any case of relapse or reinfection, and any case where any inoculation procedure has been resorted to without success, will suffice to establish the inefficacy of the method.*

(*c*) *In connexion with every cure which is credited to vaccine-therapy the possibility of that cure having occurred spontaneously must be taken into consideration.*

(*d*) *In connexion with determinations of the opsonic index, special precautions ought to be taken to eliminate the element of unconscious bias and where such special precautions have not been taken the records must be held to be suspect.*

(*e*) *In case of an author's account of what can be achieved by his method, an ample allowance must be allowed for a biased selection of cases, and for self-deception in connexion with the condition of the patients before treatment is undertaken, and in connexion with the progress they have made under that treatment.*

(*f*) *Proof of the efficacy of vaccine-therapy would be furnished only by a statistical record of the event of treatment in an extensive series of consecutive cases.*

(*a*) *Contention that the results of the blood examinations ought to be put out of sight in considering the clinical results.* This is a contention which you will find advanced, on the one hand, by those who desire that the treatment of bacterial infections should remain—at least so long as they are practising members of our profession—on the level of pure empiricism, and, on the other hand, by those who are not satisfied that there is any real correlation between the condition of the blood as revealed by opsonic determinations and the clinical condition of the patient. With the former class of objector I know that you will not, under any circumstances, have any fellowship. Nor have I any fear that you will, when you shall have studied the proof of the correlation between the clinical condition of the patient and the opsonic power of his blood, range yourselves with the latter class of objectors.

The contention that you would, if you were to shut your eyes to the record of the opsonic readings, be in a better position to judge what vaccine-therapy had done in a particular case, would then make exactly the same appeal to you as would the contention that an engineer would arrive at a better opinion on the performance of an engine if he were to close his eyes to the readings of the steam gauge.

(*b*) *Contention that nothing short of a complete cure ought to be accepted as evidence of successful vaccine-therapy, and that any case of relapse or reinfection, or any case where any inoculation has been resorted to unsuccessfully, will suffice to establish the inefficacy of the method.* This will not, I think, appeal to you as an equitable contention. You will, I think, feel that where vaccine-therapy has set out to extinguish a particular infection, and where it can be proved to have accomplished this object, this ought, from the strictly scientific point of view, to be accounted a success, even where, either in consequence of irreparable organic damage already inflicted, or of a coexisting secondary infection which has been overlooked, or of any other cause, death afterwards supervenes. Again, where a generalized infection has been held under, and the patient has thereby been restored to life and comparative health by vaccine-therapy, I think you would feel it reasonable to give the method

edit for that achievement, even if, under the influence of an inter-urrent infection, or by reason of the premature discontinuation of ne treatment, a relapse were afterwards to take place. If you are, as I eel assured, ready to place yourselves at this point of view, in adjudicat-ng upon the results of vaccine-therapy, *à fortiori* you will not counten-nce certain other palpably irrational contentions. I have in view here he contention that the method of vaccine-therapy can be discredited by he citation of a case which has been treated without success by a con-cientious and competent immunisator; and the still more egregious ontention that the method is discredited when it fails in careless and ncompetent hands. You know that success can be expected only vhere the pathogenetic agent or agents have been identified, only where y the agency of properly graduated and interspaced doses of the ppropriate vaccine or vaccines it has been possible to maintain the ntibacterial pressure at a high level, and only where the protective lements of the blood have come into operation in any nidus in which he infecting microbes may have established themselves.

(c) *Contention that in connexion with every cure which is credited to accine-therapy the possibility of that cure having taken place spontaneously hould be taken into consideration appears at first sight to be a perfectly easonable contention.* When I urge upon you that the *vis medicatrix naturae* omes insistently to the aid of the physician, even where he ignorantly pposes it, and when I ask you to reflect that among all the therapeutic nethods which have been practised there is probably not one—be that nethod never so harmful—which is not credited with many cures, you will perhaps realize that I can have no possible quarrel with the critic who lemands that it shall be demonstrated to him, in connexion with the cures which are ascribed to vaccine-therapy, that they are not to be credited to the spontaneous operations of nature.

The critic who makes this demand from me may therefore be assured that he is forcing an already open door. When he contends that spontaneous cures of bacterial disease do occur, he is only emphasizing one of the funda-mental axioms upon which the immunisator builds. For if it were not for the fact that spontaneous cures do occur, and for the fact that there follows upon these in many cases a condition of insusceptibility to further infection, the immunisator would have no warranty for expecting anything either from prophylactic or therapeutic inoculations. In other words, were it not that nature is competent to bring about these results under the stimulus of auto-inoculation, assuredly all attempts to bring them about by artificial inoculations would be vain.

But while the immunisator is in very little danger of overlooking the possibility of spontaneous recovery, there is equally present to his mind that he must, when he is considering the likelihood of spontaneous recovery, draw a sharp distinction between one kind of infection and another. Not only are there, as we have seen in the course of the dis-cussion of the expectant method, differences in this respect as between

infection by one species of microbe and infection by another, bu there is also, as was pointed out, in this respect a very fundamental dis tinction between generalized and purely localized infections. There are in other words, bacterial infections in connexion with which sponta neous recovery is the rule, others where it is very exceptional, an again others where it can hardly be said to come into consideration a all.

Now these are precisely the kind of points which are overlooked o put out of sight by the partisan objector. Exploiting, wittingly or un wittingly, the fallacy which the schoolmen spoke of as a fallacy *a dict secundum quid ad dictum simpliciter*, he would fain have you infer fron the fact that spontaneous cure is a common event in connexion witl certain types of bacterial infections, that it is to be expected in connexio with all bacterial infections without distinction.

When you have scrutinized the cases I have published of successfu immunisation, you will not fail to observe that I have limited mysel to the citation of cases which were, in my opinion, refractory or desperate cases, i.e., cases in connexion with which spontaneous cure by auto-immunisation was in my opinion improbable, or quite out of question.

We now come to the two last contentions—

(*d*) *Contention that in connexion with determinations of the opsonic index special precautions ought to be taken to exclude unconscious bias, and in particular that steps ought in every case to be taken to conceal from the laboratory worker the identity of the bloods he is dealing with.*

(*e*) *Contention that in the case of every clinical account which is given by the author of a new therapeutic method, liberal discount ought to be made for self-deception in estimating the severity of the cases before treatment, and in estimating the progress made under treatment.*

These are contentions to which no honest man can refuse to concede a certain amount of force, for bias is a factor which has to be taken into consideration as affecting to a greater or less extent every human observation.

But just as we have in connexion with the *fallacy of spontaneous recovery* carefully to distinguish between the different classes or types of bacterial infection, so here in connexion with the *fallacy of bias* we have to draw a sharp line of distinction between observations which involve a large subjective element and numerical estimations in which the subjective element practically falls out of account.

While ordinary clinical observations come under the former, opsonic determinations come under the latter category.

Let me assure you that it is practically impossible in connexion with these to practise unconscious dishonesty without afterwards finding oneself out. If, for instance, when making an opsonic count, one were, under the influence of a desire to make the estimation one was engaged upon agree with a previous one, and under the impression that one's figures were coming out too high or too low, to relax one's watch upon oneself, and

half guiltily and half unconsciously to overlook here a leucocyte and there a leucocyte which one might suspect of containing too many or too few bacteria for one's purpose, retribution would almost certainly follow in the form of too low a phagocytic count where one had sought to avoid too high a count, or in the form of too high a phagocytic count where one had sought to avoid too low a result. A similar Nemesis would dog our steps if, after counting our usual quota of leucocytes selected at random, we were to endeavour by counting a larger number of leucocytes selected under the influence of bias to alter a result which represented the correct average count of any specimen.

When we pass to the region of clinical observation we pass, as already said, to a region where unconscious bias may often mislead our judgment. It is impossible here to be sure that one might not under the influence of bias mistake a case which might readily have recovered spontaneously for a refractory case, and a case in which spontaneous recovery was not out of question for a desperate case; and that one might unconsciously exaggerate the progress that a patient made under treatment. But even here there are manifestly limits to one's power of self-deception, and we have in those cases where thermometrical measurements are available, results which can hardly be vitiated by bias.

(*f*) *Contention that definite proof of the value of vaccine-therapy could be furnished only by a statistical record setting forth the event obtained by inoculation in an extensive series of cases.*

Let me, in connexion with this contention, begin by asking you to consider what would be the evidential value of a statistical record of the event of treatment in a consecutive series of cases.

It is, I think, clear that, apart from any incidental value which it might derive from the inclusion of refractory or desperate cases, such a series of cases would acquire value only if it were placed over against a series of quite similar control cases.

Now a series of untreated cases such as would serve the purpose of controls cannot in practical life be obtained.

For such a series of untreated controls there would therefore have to be substituted, as the only possible alternative, a series of cases treated by another method, and by another practitioner. Now if this were done, the scientific issue would immediately be confused, not only by doubts as to the comparability of the two series of cases, but also by the question as to whether the therapeutic method which was applied in the control cases was hurtful, innocent, or beneficial; and above all it would be confused by a question of personal competition.

If you will consider what confusion would in this way be introduced into the issue which we are here concerned to resolve, you will, I think, understand the motives which influence me when I say that I do not propose, either here or elsewhere, to supplement by any attempted statistical proof that presumptive proof of the efficacy of vaccine-therapy which I claim to have already furnished by the citation of numerous

refractory and desperate cases successfully treated by the inoculation of bacterial vaccines.

Method of escaping from the Evidential Difficulty which would seem to arise in connexion with the circumstance that Statistical Proof of the Value of Vaccine-Therapy cannot be furnished:

I am, however, convinced that there is a very simple way out of the difficulty which would seem to arise in connexion with the fact that the presumptive proof of the efficacy of vaccine-therapy which I claim to have already furnished by the citation of refractory and desperate cases successfully treated cannot be supplemented by any statistical proof.

Assuredly my proper course is to do all I can to win proselytes and to leave it to these to furnish to you corroboration of the results which I have obtained in refractory and desperate cases.

If I have succeeded in elucidating the general principles upon which vaccine-therapy proceeds, and if I now succeed in bringing home to you that there is ample evidence of a close correlation between the results which are furnished by opsonic determinations and the clinical condition of the patient, and in bringing home to you that the method of vaccine-therapy has a very extensive sphere of application, I feel sure that there are among you scientific workers who will undertake to master the somewhat delicate technique for the measurement of the opsonic index and to carry out inoculations under that control.

I may commit it to such workers to furnish to you testimony of the value of vaccine-therapy which will be free from the fallacies which must be incident to every "author's account."

Having with this put you in possession of my thoughts, let me now try to set out the evidence which establishes that the rise and fall of the antibacterial power of the blood, and in particular the rise and fall of the opsonic power of the blood, are correlated respectively with improvement and aggravation in the clinical symptoms.

Evidence which establishes that there is a close Correlation between the Rise and Fall of the Opsonic Power of the Blood and the Clinical Progress and Regress of the Patient.

The evidence consists of data furnished by blood examination and clinical observation conjointly.

These data may be obtained by the study of the conditions which spontaneously present themselves in the course of disease, or by the study of the conditions which present themselves in patients who have been treated by bacterial vaccines. They may be obtained (*a*) *by making upon a number of patients in each case a single observation, and in conjunction with this a single measurement of the opsonic index, or* (*b*) *by following out upon patients who are kept under continuous observation in each case the events which supervene upon inoculations of bacterial vaccines or auto-inoculations.*

It will be convenient to arrange the evidence under these two head-gs, and then to consider, in connexion with the data which I furnish to ou under each of these headings, what evidential value those data might operly have for you.

vidence obtained by making upon a succession of patients in each case a single observation and in conjunction with this a single measurement of the opsonic index.

By tabulating a series of such observations made in conjunction with ouglas upon a succession of patients suffering from very chronic and rictly localized staphylococcic and tubercular lesions I have demon-rated that these were correlated with a low opsonic index with respect the pathogenetic microbe which is in question.

Vide pp. 103 *and* 118 *supra.*

By similar observations—observations which must now total up to ery many thousands—I have satisfied myself that in all bacterial infec-ons, without exception, a low opsonic index is correlated with an unsat-factory clinical condition, while a high opsonic is—with only occasional xceptions which can be very well accounted for on the hypothesis that ne focus of infection is here shut off from the circulating blood—corre-ated with a clinical condition which is for the moment improving.

Discussion of the evidential value of these data.—Assured as you re of these data only by my testimony, and unable as you are here to atisfy yourselves of the regularity with which the rule as stated above conformed to, and of the sufficiency of the explanation which is here ut forward to account for occasional departures from that rule it is lear that the data which have just been recorded can at best have for ou the value of presumptive proof of a correlation between the opsonic ower of the blood and the clinical conditions.

Evidence obtained by following out upon patients who are kept under continuous observation the events which supervene upon inoculations of bacterial vaccines.

The data which have been obtained by this means can most advan-ageously be set out in the form of "immunisation charts."

Under this heading would come (*a*) *charts relating to those localized nfections whose evolution is sufficiently acute to allow of the manifestation of objective signs of clinical progress or regress within quite short intervals.*

The charts here in question set forth in the form of a trace the changes effected by inoculation in the opsonic index, while the particulars of the clinical changes, which of course are not available in the form of quanti-tative data, are appended in the form of a verbal gloss. Charts such as these—applying as they do for the most part to infections which readily yield to vaccine-therapy—are for the most part "short charts"—charts which set forth the result of only one or two inoculations.

Those, and also the materials for such charts which have been pub-

lished by myself and by other workers, bring out (*a*) that as soon as, by th agency of the inoculation of a bacterial vaccine, the low indices whic are associated with localized infections give place to high indices, th clinical conditions improve; (*b*) that during the "negative phase" the sym toms are aggravated, and (*c*) that during the "secondary ebb" to whic attention was called above relapses are prone to occur.

For charts—or as the case may be, materials for such charts—vide supr pp. 107–111, *and pp.* 248–254.

Discussion of the evidential value of these data.—Against the evidenti value of such charts as have been in question above, the following ob jections might be urged. In the first place it might be contended tha inasmuch as the clinical data which are furnished in connexion wit these charts are furnished by ordinary clinical observation and no by any quantitative and mechanical method an allowance must in con nexion with the clinical descriptions be made for *bias*. Further, it migh be contended that inasmuch as we have here to deal only with "shor charts" the coincidences between the favourable and unfavourable change in the opsonic index and the favourable and unfavourable changes in th clinical condition might quite well be *fortuitous coincidences*. Lastly it might be urged that, inasmuch as there must in every case inevitabl be a selection of charts for publication, an element of *unconsciously biase selection* might quite well operate to bring about the publication of al the charts which showed the desired correlation and the holding back of all the charts which failed to show such correlation.

The validity of the first two of these objections so far as it concerns the charts which have just been cited can be easily rebutted. The hypothesis of bias in the clinical observations can here hardly lie, inasmuch as it is here only a question of judging whether a furuncle has got worse or has got better, and whether an existing furuncle has disappeared or a new one has made its appearance. Nor will the hypothesis of unconscious biased selection of charts lie in the case of the records here cited, for none were here excluded except on the ground other than they were not sufficiently detailed or on the ground that they were mere replicas of those published. But while these objections cannot be upheld, it will be clear to consideration that you cannot in the case of "short charts," apart from the publication of a long series of unselected charts, exclude the possibility of the simultaneous changes in the opsonic index and symptoms being the result of fortuitous coincidence.

Charts relating to generalized infections which are associated with pyrexia.—On the charts here in question are delineated two traces representing respectively the opsonic readings and the thermometrical readings. It is shown in these charts that—at any rate as soon as the auto-inoculations have been brought in some measure under control—an inverse relation between the opsonic power and the temperature readings makes itself manifest.

For charts which exhibit in connexion with the inoculation of bacterial

vaccines this inverse relation between the opsonic index and temperature,[1] *vide infra, pp.* 388–409, *Charts* 9, 10, 13, 14, 15, 16, 17, 18, 20, and 21.

Discussion of the evidential value of these data.—In view of the fact that not only the changes in the opsonic index but also changes in the clinical condition are here registered by the aid of exact quantitative measurements, and in view of the fact that in each case a sufficiently long series of inoculations is recorded to allow of the definite exclusion of the fallacies of fortuitous coincidence and unconscious biased selection of results which conform to expectation, it may, I think, without fear of contradiction, be asserted that you have here in connexion with the inoculation of bacterial vaccines conclusive evidence of a close correlation between a rise and fall of the opsonic power and a fall and rise of the temperature. Now while a rise of temperature need not invariably be indicative of an unfavourable change in the condition of the patient, and while it may upon occasion be an accompaniment of an immunising response, and while *per contra* a fall in temperature is not always indicative of a favourable change, none the less it may unhesitatingly be asserted that in a septicaemic condition a reduction of temperature is almost always indicative of an inhibition of bacterial growth in the body, and that a rise of temperature is indicative of an increase in bacterial growth. It thus follows that we have here evidence that a favourable change in the opsonic power is correlated with a favourable, and an unfavourable change in the opsonic power with an unfavourable, change in the patient's condition.

Further Evidence of Correlation.

While the charts which have just been considered constitute a very convincing form of evidence, and while they constitute the only form of evidence which is calculated to carry conviction to the critical auditor or reader who is called upon to adjudicate upon the question of correlation apart from any personal experience of the results of inoculation, it must not be supposed that these charts have any unique value for the man who has seen with his own eyes how the clinical conditions in every infection improve or change for the worse according as the opsonic index rises or falls. In point of fact, while there is not among the clinical symptoms of disease any symptom which does not vary with the changes in the opsonic index, there are among them many which bring home the fact of correlation more vividly to the observer who comes into actual contact with the cases.

It would be impossible and it is also unnecessary here to attempt anything in the nature of a complete summary of the evidence in favour of correlation which enforces itself upon the notice of the immunisator as

[1] *For charts which show in connexion with auto-inoculations instead of this inverse relation a direct, and up to the present unexplained association between a rise and fall of temperature and a rise and fall of the opsonic index, vide pp.* 387–388 *and* 391–392, *Charts* 7, 8, 11, and 12.

he day by day compares the results of his blood testings with the clinica condition of his patients. Brief reference may, however, be made t some of the more striking evidence.

The fact that the favourable issue of pneumonia is associated wit a rise in the opsonic index is brought out very clearly in the charts whic have been published by MacDonald [1] from Dr. Bulloch's laboratory. I is here shown that during the course of pneumonia the opsonic power i continuously subnormal, and that the *crisis* in pneumonia occurs i connexion with a sudden and striking rise in the opsonic power of th blood. The same association between a rise in the opsonic index—a rise here achieved as the result of the inoculation of a pneumococcus vaccine—and clinical improvement in a pneumococcic infection is shown by Eyre. It is again shown by the workers in Hektoen's laboratory that the favourable or unfavourable event of pneumonia is correlated with a rise or fall in the opsonic index.

The fact that changes in the opsonic index stand in close relation to increased or diminished *pain* comes strikingly under observation in connexion with the treatment of gonococcal rheumatism by gonococcus vaccine.

Chart 6, p. 386 infra, sets forth in a very striking manner the close correlation which exists between the clinical symptoms and the readings of the opsonic index in gonococcal rheumatism.

The same association as in gonococcal rheumatism comes under observation in connexion with the treatment of *tubercular arthritis* by tubercle vaccine, the conditions being here only so far different that we have here to deal, not so often with pain, as with a feeling of weakness and of fatigue in the joint.

The fact that changes in the opsonic index stand in close relation to increased or diminished pain comes again under observation (it comes here under observation in association with increased or diminished *frequency of micturition*) in *tubercular cystitis*. So marked is this association that it is in the ordinary case possible before the blood is tested to foretell from the symptoms whether the opsonic index will turn out to be high or low.

A very striking association between the severity of the *pain* and the opsonic index comes under observation again in the case of patients who are the victims of *malignant disease*. Here, in practically every case, the pain of the associated inflammation is relieved when, by the inoculation of appropriate doses of a vaccine made from the staphylococcus which was denoted by Doyen *micrococcus neoformans*, the opsonic index to this micro-organism has been raised. And the pain as regularly recurs when the opsonic index to this micro-organism again falls.

A similar correlation between the amount of purulent discharge and the opsonic readings comes under observation in connexion with purulent discharges from mucous membranes—in particular in connexion with gonococcal discharges.

[1] *Transactions Pathological Society of London*, 1905.

Charts showing the correlation which exists between the opsonic readings oth in gonorrhoea and otitis are set forth in the papers of Hektoen's upils.[1]

A correlation, and a very striking correlation, between the *psychical ymptoms* and the readings of the opsonic index may be observed in a arge variety of bacterial infections. Low indices in tuberculosis are or the most part associated with low spirits; high indices with high pirits. The *spes phthisica* may very probably be associated with the mmunising responses with which the organism responds to the tubercular uto-inoculations. In a very similar way in the case of coli infections he patient's outlook upon life varies with his opsonic index.

Finally, in connexion with chronic and long-lasting infections, such as tubercle, it may be observed that where the disease is in process of being completely extirpated, the opsonic index maintains itself after noculation for considerable periods at a level well above the normal *maintained high tide of immunity*), whereas in cases where the infection s not yet being satisfactorily overcome, the index always falls away apidly to a subnormal level.

Let me now, with a view to showing you how extensive is the sphere of application of vaccine-therapy, try to summarize for you my personal experience of the results of vaccine-therapy. Let me deal separately vith each of the more important types of infection.

Summary of the Results which have been obtained by Vaccine-therapy.

Strictly localized infections affecting the subcutaneous tissue, lymphatic glands, bones, joints and other parts of the body.

Typical examples of this class of infections are to hand in the case where staphylococci or streptococci have penetrated into the subcutaneous tissue, and in the case where tubercle bacilli have effected a lodgment n lymphatic glands.

In these cases, and in the whole class of cases of which these are examples, all but uniformly successful results are achieved by vaccine-therapy supplemented, as occasion has required, by measures for determining a free lymph flow in the focus of infection.

In the case of ordinary furunculosis improvement has generally been almost immediate. On the other hand, in cases where the tissues have been much infiltrated, and where—as in carbuncle—the lymph flow is obtruded, improvement has never been rapid until a free flow of lymph has been established in the affected region.

In the case of tuberculous infection of the lymphatic glands the period of treatment—measured from the inception to the complete retrocession of the swelling—has varied, according to the extent of the infection and the individual patient's power of immunising response,

[1] *Centralblatt f. Bakteriologie.* I. Abth. Orig. Bd. xliv, Heft 5.

between five weeks and eighteen months. It has, on the whole, average about six months. Where suppurating tuberculous glands have bee in question, or where suppuration has supervened in the course of th vaccine treatment, this complication has in every case been successfull dealt with by vaccine-therapy after incision, or aspiration, supplemente by chemical suction.

What applies to tuberculous infection of the lymphatic glands applie in a general way also to tuberculous infection of the testicle, and t simple tuberculous infection of the kidney and urinary passages.

It applies also—but on this question I speak with a reserve impose upon me by a very restricted experience—to early cases of tubercle o the lungs.

Ulcerative Type of Infection.

In my experience this type of infection—a type which is met wit in connexion with the breaking down of nodules in the deeper tissues and in connexion with the invasion of those tissues from the sur face—does not differ with respect to its tractability to vaccine-therap from the type of infection last considered, except only in the cas where secondary infections have supervened. If anything—given th case where secondary infections have either been avoided or been suc cessfully combated—an open ulcer is more tractable to vaccine-therap than a focus of infection in the deeper tissues which has not yet foun external vent, and more tractable than a focus of infection in the epidermi which has not yet penetrated to the underlying lymph-bearing strata It will be clear that, as soon as the subcutaneous tissues have been tappe the lymph stream which courses through these will well up throug the floor of the ulcer, coming into application, as it does so, upon th infecting micro-organisms.

Infections of the Skin.

Infections of the skin fall naturally into two categories. Wher the infected skin is comparatively dry and scaly and non-vascular w are dealing with a form of infection which is, in my experience, relativel intractable to vaccine-therapy. A typical example of such a type o skin infection is furnished by the superficial scaly form of lupus whic has from the point of view of its superficial resemblance to psoriasi been, very appositely, denoted "*lupus psoriasis.*" Where the microbe penetrate deep into the skin we have forms of infection which ar very tractable to vaccine-therapy.

Infections of Mucous Membranes and of the Glands and Ducts which stand in connexion with Mucous Membranes.

Infections of mucous membranes are, in my experience, very readil influenced by vaccine-therapy. Many very successful results have been obtained in connexion with the most various infections of the middle ear, antrum, nasal sinuses, dental alveoli, and salivary glands. Successfu results have also been obtained in connexion with coli infections of the

ıtestinal mucous membrane and gall-bladder. The same holds true ı connexion with many different infections of the uterus, kidney, urinary ladder, and urethra.

It is to be noted in connexion with many of these infections that hey have been mixed infections, and in connexion with the infections f the urinary passages in particular that there was often superadded o the infection of the mucous membrane a bacteriuria. The extinction f this bacteriuria—which is an incomparably more difficult task than he relief of cystitis and the extinction of the infection in the mucous nembrane—has been achieved in a considerable percentage of the ases which I have treated.

Infections of Sinuses.

In my experience very successful results are obtained in these cases vhen the inoculation of bacterial vaccines is combined with the application of a local lymphagogue.

In conclusion, I may perhaps say a word on the subject of results hat have been obtained in the treatment of "mixed infections," and n the treatment of "generalized infections."

Mixed Infections.

While the suggestion that mixed infections must be expected in suppurative processes occurring in connexion with surfaces which harbour nicrobes may quite well be universally acceptable, as not breaking in ıpon any accepted belief, the suggestion that the question of mixed nfection must perforce be considered in connexion with every case of phthisis, lupus, tubercular caries, tuberculous cystitis, and tuberculous ılceration will, in the very nature of things, be unacceptable to many clinicians. Such a suggestion will be felt to call in question both the clearness of vision of those who in connexion with these diseases clamour, and have clamoured, for antituberculous remedies only, and the critical acumen of those who, without taking into account the fallacies which are incidental to clinical methods, confidently undertake to adjudicate on antituberculous remedies by the naked eye observation of the effects upon cases where, in addition to the tubercle bacillus, other pathogenetic microbes are at work.

Be it acceptable or unacceptable, there is no escape from the fact that practically every case of suppurating lupus is complicated by staphylococcus infection, and that the majority of aggravated cases of lupus are complicated by a streptococcus infection.

What holds true of lupus holds true of the majority of tubercular sinuses.

Having appreciated the magnitude and the far-reaching nature of the issues involved in the treatment of mixed infections, we may come to the question of the results achieved in these cases by vaccine-therapy. We have two cases to consider.

Case where vaccine-therapy is directed to the destruction of only one of the different species of infecting microbes.—In a few instances—notably

in two cases of rupial furunculosis where we had to deal with a mixtur of streptococci and staphylococci—the extinction of one of the microbe under the influence of the corresponding vaccine has indirectly led t the extinction of the other. This event is, however, extremely excep tional.

In most cases the employment of vaccine-therapy directed to th destruction of a single species of microbe leaves the other species quit unaffected. It may even—and this applies in particular to surfac infections of mucous membranes or ulcers—leave the ground free for th multiplication of the other—i.e., the originally competing microbe.

Case where vaccine-therapy is directed to the destruction of each of th different species of infecting microbes.—Where in cases of mixed infectio measures are taken to immunise the patient against each of the differen infections very successful results have been achieved. Successful result have been achieved, notably in the case of lupus, cystitis, and endome tritis. While naturally the task of the immunisator is more laborious an more intricate in the case where two or three different vaccines are em ployed, it would seem that the organism of the patient does not fin the task of responding to a number of different vaccines (always sup posing that each of these is administered in appropriate and properl interspaced doses) given together more difficult than the task of respond ing to one variety of vaccine only.

Generalized Infections.

In association with my fellow-workers I have, up to the present, treate by vaccine-therapy some half-dozen cases of Malta fever and an equa number of cases of streptococcal septicaemia.

In each of the cases of Malta fever the course of the disease would seem to have been favourably influenced—the clinical improvement occurring in each case in association with an increased development o antibacterial substances in the blood.[1]

In the cases of streptococcal septicaemia [2] the results have been as follows :—

In two cases—one of these being a case of malignant endocarditis —a complete cure was achieved, in each case in association with a very satisfactory immunizing response. In a third case—also a case of malignant endocarditis—the high temperature which had lasted for three months before vaccine-therapy was resorted to, came down to the normal under the influence of the inoculations, the patient making an excellent immunising response. In this case death by cardiac complica- tion occurred on the fourth day after defervescence.

In three other cases of streptococcal endocarditis the patient suc- cumbed, having in each case failed to make any immunising response to the inoculations.

[1] *For a Chart applying to one of these cases, vide infra, Chart* 13, *p.* 389.
[2] *For details and Charts of these cases, vide infra, pp.* 388–405, *Charts* 13–21.

VIII.—Concluding Remarks.

I would venture in conclusion to come back once more upon the erapeutic measures which are currently employed for the purpose of mbating bacterial infections. I want to come back upon them, in der to consider in connexion with these and with vaccine therapy e further point. If disillusion and disappointment have followed on the enthusiastic exploitation of antiseptics, surgical extirpation d serum-therapy, we may well ask ourselves whether in the future e same tale will not have to be told of vaccine-therapy.

To this there is but one inevitable answer. Unless we can detect, and less it should be given to us to avoid, the fallacies which have led to justified expectations and consequent disillusion in connexion with rrent therapeutic methods we shall assuredly not escape such disillusion connexion with any newly introduced therapeutic method.

Our sins of omission and commission in connexion with the thera- utic methods which were mentioned above, were, I apprehend, the llowing :—

(1) *Sufficient thought was not in any case devoted to the problem which s to be attacked.* In the case where antiseptics were employed to mbat bacterial infection, the issue as to whether the conflict of the ganism with the infecting microbe might not be paralysed by the gestion or application of the antiseptic was in practice overlooked. was the issue as to whether anything in the nature of a complete rilization could by these means be achieved. In connexion with surgical tirpation the issue as to whether it is theoretically possible to extirpate th the knife all the infecting micro-organisms was put out of sight. the case of serum-therapy it was assumed without either *à priori* stification or experimental proof that the animal which was vicariously oculated would respond to any kind of demand that might be made on it by the immunisator.

(2) *It was not in connexion with any of these therapeutic methods thought cessary to study in a systematic manner by bacteriological investigations e effect of the treatment upon the infecting bacteria and the patient's power resistance.*

I may for instance point out to you that it has not been thought neces- ry or worth while in connexion with any form of serum-therapy to dy on the patient the effect of that treatment upon the antitoxic or tibacterial power of the blood.

(3) *We permitted ourselves in connexion with each newly introduced rapeutic method to indulge in extravagant expectations.*

In reality, far more important consequences than one might at first ppose depend upon a proper regulation of expectation in the field of dicine. So long as we as a profession go on hankering after the impos- le, so long as we demand of every new therapeutic method that it all after the manner of a magic wand achieve the marvellous with

little labour, and that it shall give its best results even when it is applie
in an absolutely blind empirical manner, so long will disillusion continu
to dog the steps of medicine.

There will be an end, once and for all, to such clamouring for the moo
as soon as science shall have imported order and measure into our expec
tations. It will then come home to us that when man has in any branc
of applied science obtained results commensurate with the knowledg
and labour which he has brought into application he has obtaine
everything he is entitled to ask or expect.

Now in medicine we have aspired to directive control over bacteria
infections without thinking it necessary to acquire an intimate knowledg
of the phenomena of infection, or of the consequences of intervention
or of the *modus operandi* of our remedial agencies.

Inherent Limitations of Vaccine-therapy.

It would therefore be well that in connexion with vaccine therapy w
should appreciate the limitations which are inherent in the method, and th
labour which its proper conduct may entail, and further that we shoul
distinguish between the cases where we may hope to achieve certain an
easy success, and the cases where success, if achieved at all, must be dearl
bought.

(*a*) We must keep constantly in mind that where we have recourse t
vaccine-therapy we furnish to the organism not the protective substance
which are required, but only a stimulus which will under favourabl
conditions elicit a production of such substances. It is an obvious corol
lary that the success of the treatment is in every case conditional, o
the one hand, upon the right choice, and right dosage, and right inter
spacing of the vaccine, and, on the other hand, upon the patient's indivi
dual capacity for immunising response.

(*b*) We have further to call to mind that the protective substances whic
are formed in response to the inoculation of a bacterial vaccine disap
pear in the infected organism rapidly from the blood.

It follows that if we are not immediately successful in destroyin
all the infecting micro-organisms—and such immediate success ca
hardly be expected except in the case of incipient infection—we hav
to make up our minds to go in for a programme of periodical inoculation
—just as a gardener who undertook the task of clearing the weeds out o
a neglected garden would make up his mind to renew his efforts week b
week until his purpose was accomplished. And just as in gardenin
no one finds any ground for surprise if the weeds spring up again
when a residue has been left in the soil, or when the ground lies ope
to seeds borne in from without, so in connexion with bacterial infection
recrudescences and fresh reinfections must be expected where a residu
of microbes are left in the body, and where the channels through whic
infection originally found entrance are left open and unguarded.

(*c*) Where we are dealing with generalized infections or localized infec

ons which have reached the stage at which auto-inoculations are ccurring vaccine-therapy encounters special difficulties.

(*d*) Lastly, we have to realize that what can be directly achieved by accine-therapy is nothing more than a greater protective power in he circulating blood. Hence when we aim at the destruction of nicrobes which are as it were barricaded round in the tissues we can-ot expect success until those barricades have been broken through.

Vhat Bacteriological Training and what amount of Bacteriological Labour is required for the Proper Treatment of Bacterial Infections by Vaccine-therapy?

When as in ordinary cases of furunculosis, or lymphangitis, or erysipe-as, the clinical appearances immediately tell us the infecting microbe, nd the required vaccine, and when the proper dosage of the vaccine for uch cases has already been worked out and made known, treatment by accines, like the empirical use of proprietary medicines and drugs, may e undertaken, and for the most part successfully undertaken, without nything in the nature of special knowledge, and with no more technical kill than is required for administering an injection of morphia.

If all bacterial infections were as tractable as those just mentioned, nd if the appropriate dosage of every variety of vaccine had been as ully worked out, and, above all, if there was any such thing as an optimum ystem of dosage which would apply to all cases alike, or if in all cases he condition of the patient was such that it was a matter of indifference s to whether we obtained the best attainable results, or something less han the best results, then vaccine-therapy might be reduced to a mere ule of thumb.

But in Nature nothing of this kind ever does work out as a system of ule of thumb, and just as in technical chemistry and engineering the rule of humb artizan has been supplanted by the scientific craftsman, so also vill it be in medicine. Hence the real issue we have to discuss here is not he question as to what is the irreducible minimum of medical knowledge vhich is required for vaccine-therapy, but rather what is the minimum of acteriological knowledge and technique which must be at disposal in rder to ensure that the results which are obtained shall in every case be he best which are at present obtainable.

Manifestly the immunisator must for the purposes of diagnosis and reatment have at his command whatever knowledge and technique nay be required for identifying the microbes in both " open " and "closed" nfections, for gauging the gravity of each infection, for determining vhether a dose of vaccine is too large or too small, for ascertaining whether he time has arrived for reinoculation, and for satisfying himself—when s the result of treatment the overt symptoms of the disease shall have lisappeared—whether the infection has really been completely extin-guished.

For the identification of the more common and more formidable and

more easily recognized pathogenetic microbes in "open infections, a training in bacteriology and technique such as may under favourabl circumstances be acquired by students in an ordinary course of bacterio logy ought perhaps to suffice. But for all the other purposes enumer ated such training is hopelessly inadequate. It does not impart, it doe not even aim at imparting, that intimate acquaintance with the bacterio logy of the human body in health and disease which is the first require ment of the physician who seriously intends to diagnose bacteria diseases and to apply to each its specific treatment. And again th ordinary student's course of bacteriology does not—any more than doe the ordinary student's course in chemistry—impart any training i accurate quantitative work.

In both the one and the other of these respects—and I trust I ma say so unblamed—the education of even the professed bacteriologist i at fault. On the one hand, unless he has given special study to thi subject, he has not at command that knowledge of the bacteriology o the human body which an immunisator requires, and, on the other hand his rare attempts at quantitative work—consisting as they do of littl more than the enumeration of colonies on Petri dishes, the making o dilutions in more or less cumbrous ways, and the measurement of bacteri by loopfuls, or where larger quantities are in question, by fractions o multiples of agar tubes or bouillon cultures—all these attempts, I say bear much the same relation to quantitatively accurate bacteriologica methods as the technique of the beginner at billiards bears to that of a professional.

I must therefore insist that a serious discipline in quantitative tech nique—a discipline similar to that which is required before any one can play billiards or do any effective work in chemistry—is an indispensable preliminary to undertaking quantitative bacteriological work for the pur poses of diagnosis or guidance in immunisation.

I pass from the discussion of the bacteriological training which is required as a preparation for the work of an immunisator to the question of the amount of bacteriological labour which will be called for in con nexion with each particular case.

We may here consider work (1) in connexion with *diagnosis*, (2) in connexion with the *preparation of vaccines*, and (3) in connexion with the *regulation of the treatment.*

(1) *Work in connexion with diagnosis.*—In connexion with diagnosis we have three types of cases to consider: (*a*) cases where the nature of the infection is obvious from the symptoms, (*b*) cases of "open infection" where the nature of that infection is uncertain, and (*c*) cases of "closed infection," where the nature of that infection is uncertain.

It is clear that no diagnostic work of a bacteriological nature is called for in connexion with the first class of cases.

In the second class of cases microscopic preparations and cultures must be made from the discharges, and in the case where these methods

ive negative results, or where we suspect that in addition to the nicrobes which are found there may be other microbes which are un-epresented in the films and cultures, inoculations into animals must be employed, or we must resort to those methods of inferential diagnosis which are applicable in the case of "closed infections." Moreover, it must be kept in view that, however exhaustive our initial diagnostic examination of an open infection, we cannot rest content with this. The diagnosis must from time to time be revised, for new varieties of microbes may invade the wound, and varieties originally present may disappear.

In the case of "closed infections" direct methods of diagnosis are of course inapplicable. Here a series of measurements of the patient's opsonic index must be made with a view to determining whether his index with respect to each suspected variety of micro-organism fluctuates spontaneously, or can be made to fluctuate by activating the lymph stream in the focus of infection.

(2) *Work in connexion with the preparation of vaccines.*—With respect to this it will suffice to say here that while it would seem probable that in all cases the best results would be obtainable by the use of vaccines prepared from the individual patient, and while such vaccines ought, wherever possible, to be resorted to in dangerous or obstinate infections, in the ordinary case, stock vaccines give very satisfactory results.

(3) *Work in connexion with the regulation of the treatment.*—We have again three cases to consider (*a*) the case of comparatively light infections where the symptoms give us adequate guidance in the regulation of our inoculations, (*b*) the case of localized infections where the symptoms afford no guidance in the treatment, while at the same time our proceedings are not interfered with by auto-inoculations, (*c*) the case of generalized infections where we have to reckon with irregularly recurrent spontaneous auto-inoculations.

As in the *first* case considered in connexion with diagnosis, so here in the first case which has to be considered in connexion with treatment, bacteriological work may be dispensed with.

In the *second* case it will generally suffice if we inform ourselves from time to time by blood examinations that our scheme of dosage is correct. But where more rapid and certain results are required we shall do well to control each dose, testing the blood in the manner already explained before and twenty-four hours after each inoculation. And where the progress of the case leaves something to be desired, more frequent blood examinations, or even daily blood examinations may be advisable.

In the *third* class of cases, where we are dealing with an irregular and almost continuous series of auto-inoculations, it will be imperative, if we propose to intervene by inoculation, to keep in touch with what is happening in the body. This involves measuring the opsonic index once a day, if not oftener.

On consideration of this onerous programme of work you will inevitably ask yourselves whether it would not be possible to substitute for the

measurement of the opsonic index some unlaborious clinical observation or whether, in default of this, it would not be possible to substitute for the measurement of the opsonic index some less laborious method of blood examination.

With regard to the former question I would emphasize that there is no clinical observation which can be accepted as an equivalent to a measurement of the opsonic index.

In connexion with the latter question I would point out, on the one hand, that there is no other available method, and, on the other hand, that, as experience in connexion with every other branch of science teaches, quantitative determinations are of necessity laborious.

Having now come quite to the end of my task, let me add one final word on a question whose importance will be manifest as soon as I have formulated it for you.

Question what Cases give for the Labour entailed upon the Immunisator the largest Return of Advantage to the Patient.

The wholly unscientific atmosphere which pervades almost all our medical thinking shows itself perhaps most conspicuously in this, that it hardly occurs to you and me to begin in medicine with the simple and easy problems, and to advance from these to the more complicated and difficult ones.

Setting our hearts upon the immediate discovery of, shall I say, a cure for cancer, or for advanced consumption, or for septicaemia, we fail to reflect that in the case of the first we have not yet found that loose end which, followed up, might perhaps lead to the unravelling of the problem, that in the case of the second we have probably a multiform mixed infection complicated with what are in effect voluminous open abscesses, and that in the case of septicaemic diseases we have to deal with the most uncontrollable types of infection.

And, aspiring after a remedy for these, we have neglected to work at the comparatively simple and soluble problems of localized diseases.

We must wholly put away from us all such immoderate ambition if we are in connexion with vaccine-therapy to reap the best results from our labour.

Except only when we are striving to open up new fields we shall do well to embark with the greatest reluctance upon the treatment of very grave septicaemic cases, and to reserve our working strength for really fruitful work in connexion with localized and incipient infections.

Studies in Connexion with Therapeutic Immunisation.[1]

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—I. Charts illustrating Points in connexion with the Immunising Reaction which is obtained in Response to the Inoculation of Bacterial Vaccines—II. Charts showing Auto-inoculation Effects in connexion with the First Beginnings of Tuberculosis Infection—III. Chart showing in connexion with a Case of Gonococcal Arthritis, Auto-inoculation and Immunising Response, and the Correspondence which exists between these Phenomena and the Clinical Events—IV. Charts showing Auto-inoculation Effects in connexion with Advanced Phthisis—V. Charts showing Auto-inoculation Effects in connexion with Generalized Infections—VI. Charts showing (*a*) the Conditions which are met with in Man in Streptococcal and other Septicaemias, and (*b*) the Results which have been obtained in these Conditions by Recourse to Vaccine-therapy—VII. Charts illustrating the fact that Auto-inoculation Effects are obtained where Active or Passive Movements are undertaken in connexion with Localized Bacterial Infections—VIII. Charts showing that Auto-inoculation Effects are Elicited by Operative Interference with Foci of Bacterial Infection—IX. Charts showing that Auto-inoculation Effects are obtained by Active and Passive Hyperaemia affecting Foci of Bacterial Infection.

I.—Table illustrating some of the Diagnostic and Therapeutic Problems that can be resolved by recourse to the auto-inoculation test.

Ve have in the course of the past three years in connexion with the reatment of out-patients in the Department for Therapeutic Immunisa-ion at St. Mary's Hospital, of in-patients in the Hospital, and of patients outside the Hospital, had opportunity of investigating a number of points n connexion with the diagnosis of bacterial diseases and their treatment by methods of immunisation. We reproduce below a selection of some of the more interesting of our records.

We have divided our paper into two parts.

In Part I we treat first of certain unregarded points in connexion with the immunisation curves which are obtained by bacterial inoculations. We then deal with auto-inoculations, showing that these may come under observation in connexion with the first beginnings

[1] Reprinted from the *Lancet*, November 2, 1907.

of tuberculous infection, and that they are a regular accompanimen of the hectic fever of advanced phthisis. We then show in con nexion with a case of gonococcal arthritis that there is a very inti mate relation between auto-inoculation and auto-immunisation on th one hand, and the clinical symptoms of the patient on the other. W then take up the question of generalized bacterial infections and show that spontaneous auto-inoculations and immunising responses are characteristic feature in anthrax septicaemia as seen in rabbits. From thi we pass on to deal with human streptococcal and staphylococcal septi caemias, setting out here all the work which we have done with a view to eliciting immunising responses by the agency of vaccine therapy in those cases where spontaneous immunising responses make default. Finally we set forth some of the more interesting of the records which we have obtained in the course of systematic study of auto-inoculations in con nexion with localized bacterial infections. We here consider, first, the effect of massage and of active muscular movements affecting the focus of infection ; then the effect of operative interference with such foci ; and lastly, the effect of active and passive hyperaemia affecting these foci. The forty-four records which we bring forward in illustration of these various points are all submitted to the reader in the form of charts provided with a brief explanatory commentary.

In Part II we bring forward evidence to show that we have in the induction of an auto-inoculation, when this is preceded and followed up by a series of measurements of the opsonic index, a method which can be turned to account for the resolution of some of the diagnostic and therapeutic problems which present themselves for solution in connexion with every localized infection which is not accessible to direct bacteriological examination. Of the fifty-one records which we bring forward in illustration of the problems which can be resolved by this method one is, by way of illustration, submitted in graphic form. The remaining fifty are for convenience of study arranged in a table.

PART I

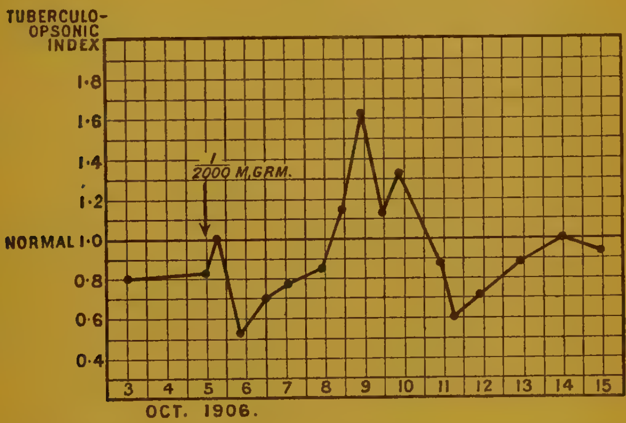
I.—Charts illustrating Points in connexion with the Immunising Reaction which is obtained in Response to the Inoculation of Bacterial Vaccines.

Chart 1 exhibits, in addition to the negative and positive phases, which are already familiar features in connexion with every immunisation curve, two minor features which are not without a certain interest. The first of these is the upward movement of the curve which follows directly

oon inoculation anticipating the negative phase.[1] The second is the nking away of the curve after the positive phase to a point below the riginal point of departure.[2]

The first of these features is of interest as explaining the immediate inical improvement which not infrequently supervenes directly upon ne inoculation of a bacterial vaccine. The second is of interest as ex- laining the fact that indices appreciably lower than those encountered n the untreated condition may be met with subsequent to the inoculation f vaccines. To be noted in connexion with the secondary ebb of the urve of immunisation is the fact that the point of lowest ebb has no

CHART 1.



Owing to the fact that the quantum of dry tubercle powder in Koch's T.R. was declared by them as ten milligrammes in one cubic centimetre, when it was in reality only two milligrammes in one cubic centimetre, the dose here inoculated would in reality be only $\frac{1}{10000}$th milligramme.[3]

sooner been reached than—and this is seen also in Charts 3 and 6— the tide spontaneously turns again.

Chart 2 exhibits the results obtained by measuring the agglutinating power of a guinea-pig which was subjected to successive inoculations of a vaccine consisting of a culture of the glanders bacillus which had been sterilized by heating to 60° C. It will be seen that we have here on record in connexion with the agglutinating power of the blood a succession of negative and positive phases exactly comparable to the succession of negative and positive phases which are registered where, in connexion with bacterial inoculations, successive measurements of the opsonic or,

[1] This feature is again exhibited in Charts 27, 30, 38, 39, 41, and 44, and again in the Table under serial numbers 6, 10, 13, 17, 27, 28, 29, and 31.

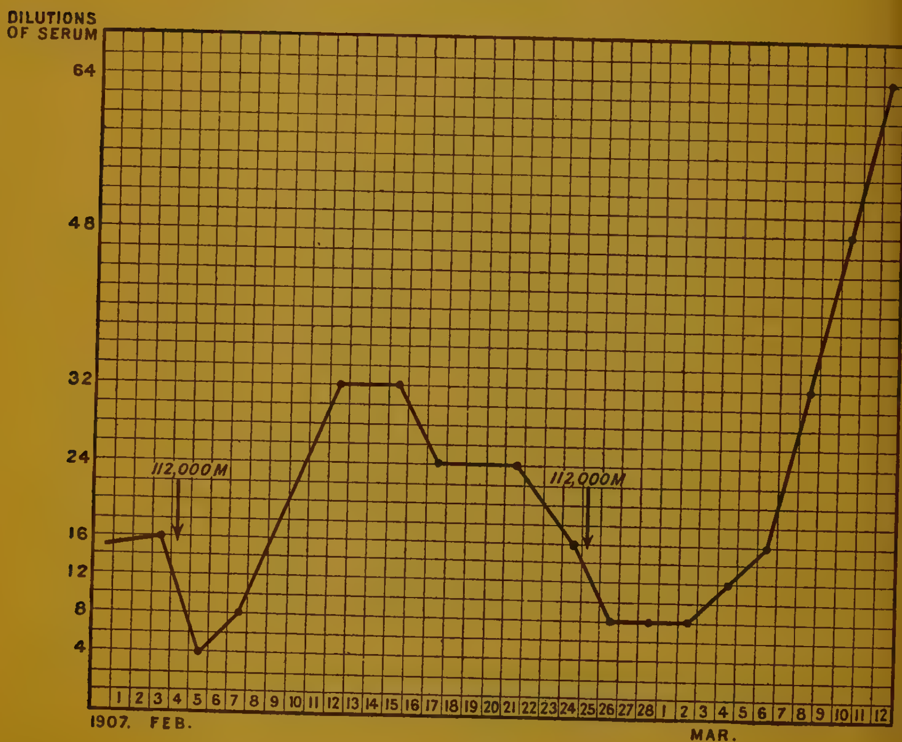
[2] This feature is again exhibited in Charts 3, 5, 6, 23, 26, 30, and 40, and in the Table under serial numbers 14 and 28.

[3] *Vide supra*, page 123, footnote No. 2.

as the case may be, of the bactericidal power of the blood are undertaken. It is thus clear that the course of the immunising reaction can be followed by measurements of the agglutinating power of the blood.

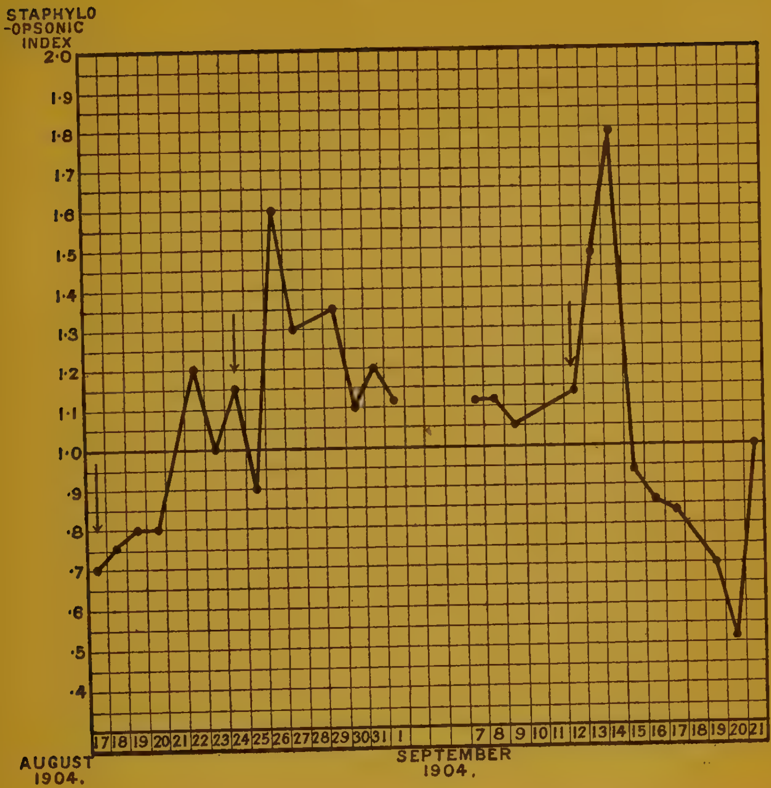
Chart 3 refers to a laboratory boy who was the subject of acne and who was inoculated by us with staphylococcus vaccine. To be noted in

CHART 2.



connexion with the chart is (*a*) the fact that the first two doses of vaccine gave a cumulative effect in the direction of the positive phase; (*b*) the fact that there supervened upon the very high "spring-tide" which was evoked by the third inoculation a very pronounced "secondary ebb"; and (*c*) the fact that the progressive improvement which was achieved in association with the positive phases which are here on record was, in the period of the secondary ebb, succeeded by a temporary aggravation of the patient's condition.

Chart 3.



II.—Charts showing Auto-inoculation Effects in connexion with the First Beginnings of Tuberculous Infection.

The two following charts which refer to past workers in this laboratory appear to us to possess quite exceptional interest in connexion with the detection of the first beginnings of tuberculous infection. Before considering them we may premise the following : (*a*) In connexion with the measurements of the tuberculo-opsonic index which are carried out in this laboratory, as a matter of daily routine, we employ as controls the sera of from two to four laboratory workers, with respect to whom we feel confident that they are not the subjects of any tuberculous infection. Both the laboratory workers whose charts are here in question came under this description. (*b*) In the ordinary case where the blood of a patient is in question we work out the opsonic index by dividing the phagocytic count which we obtain with his serum by the mean of all the phagocytic, counts which we obtain with the control sera. In the case of the laboratory workers here in question the index was in each case obtained by dividing

the phagocytic count corresponding to the particular worker's serum by the mean of the phagocytic counts of himself and his fellow-workers.

CHART 4.

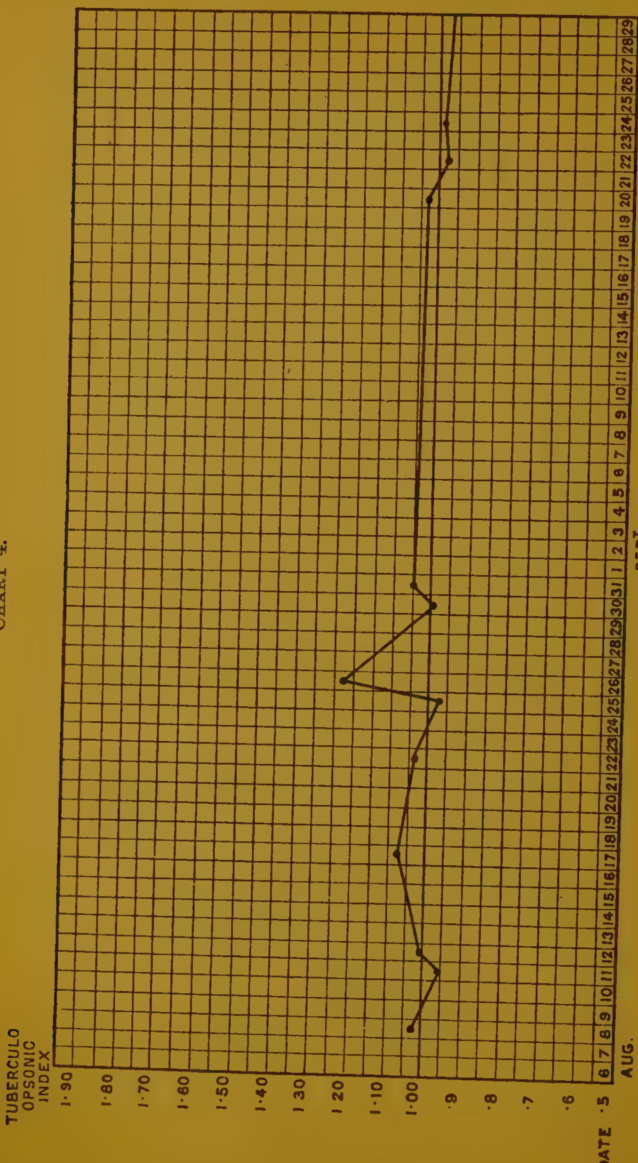
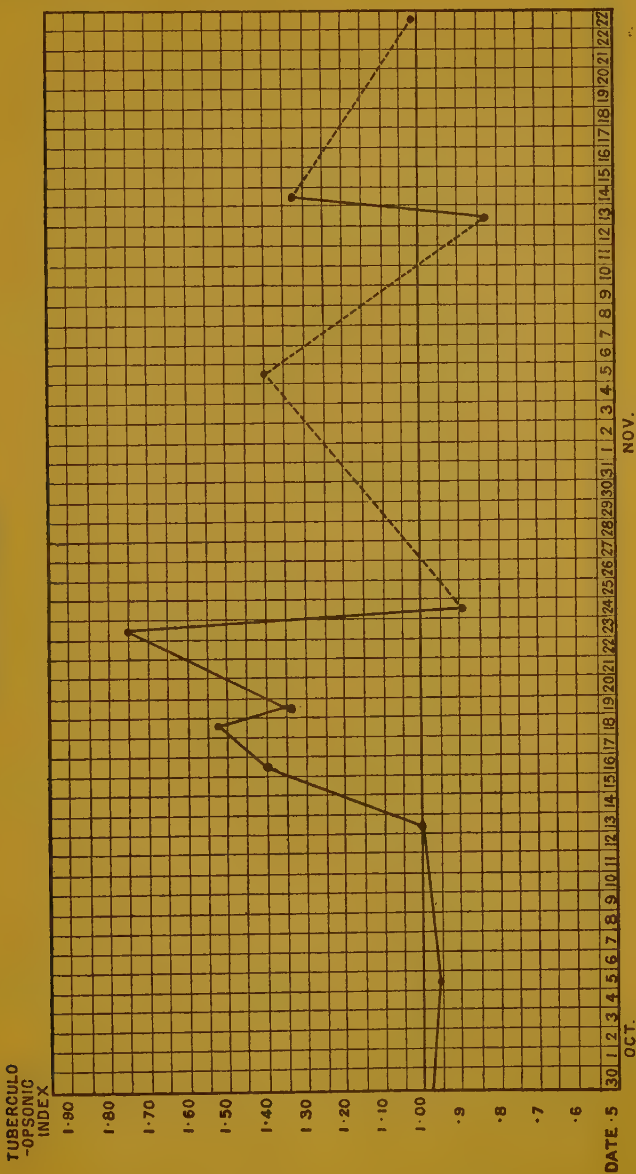


Chart 4, which refers to the first of the two cases which are in question, shows that during the ten weeks which are covered by the first two-thirds

the chart the tuberculo-opsonic index of our fellow-worker's blood
ver ranged below 0·9 or above 1·1, except on a single occasion (in

CHART 4 (continued).



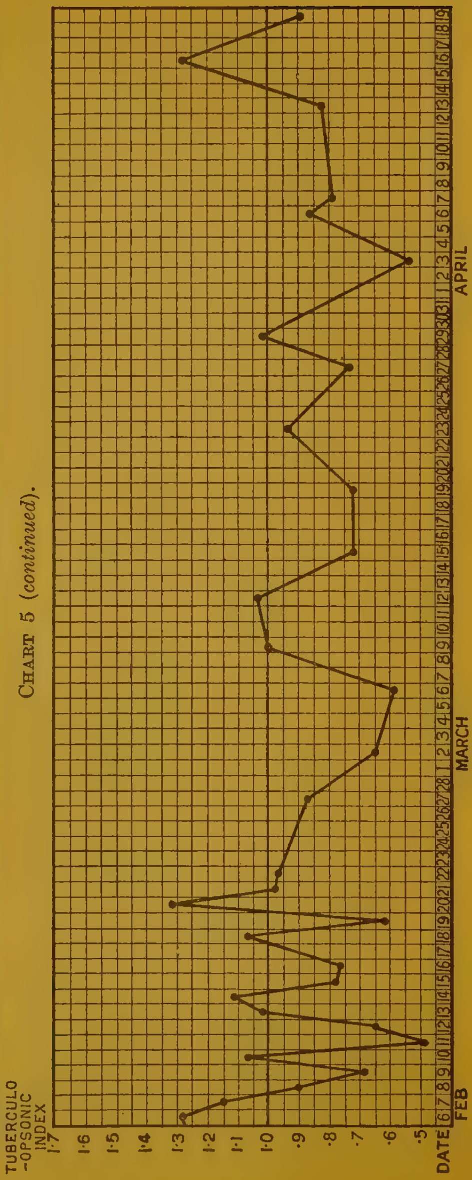
ugust) when a reading of 1·25 was obtained. In the middle of October,
or the first time, a serious discrepancy made itself manifest between this

worker's blood and the other control bloods. With a view to probing thi matter to the bottom we took samples of blood from each of the worker in the laboratory—then some ten in number—and tested each individua worker's serum against the pooled serum of all the workers. It immedi ately emerged that the serum of the particular worker whose chart is her in question showed the abnormally high tuberculo-opsonic index whic signalizes an immunising response to a tuberculous inoculation or auto inoculation. An acute febrile attack now supervened. It was diagnose as "typical influenza." Our fellow-worker, who was a man of splendi physique, however, lost flesh very rapidly, developed a cyanotic hue and complained of a certain amount of tenderness in connexion with th lymphatic glands of the neck. In association with these symptoms tuberculo-opsonic reading of 1·75 was obtained, and twenty-four hours afte a reading of 0·9. It was now ascertained that the patient had more tha ten years before undergone an extirpation operation for tuberculou glands in the supra-clavicular region. We take it, in view of all thes circumstances and of the subsequent events, that we were here face t face with what we are persuaded must be a comparatively commo event—to wit, a tuberculous septicaemia.

Of quite special interest is the subsequent history of this case. Th patient made a rapid convalescence, recovered the weight that he ha lost, and returned to work in the laboratory. His tuberculo-opsoni index none the less showed the fluctuations delineated in the las quarter of the chart. To us it seemed clear that we had here evidenc of tuberculous infection. To others the patient's appearance mad such infection quite incredible. To believe here in tuberculou infection, and to believe in it upon the evidence of a fluctuatin opsonic index alone might quite well, so they reasoned, be fruit of a min diseased. Our fellow-worker none the less elected to exchange the lat hours and the physical strain of laboratory work for an outdoor life. H reported to us before long that the fluctuation of his tuberculo-opsoni index had not been without significance. He had developed tuberculou epididymitis.

Chart 5 refers to a laboratory worker who took the place of the worke last in question. Here we have:—in the last week of November and th first week in December, nine successive tuberculo-opsonic readings whic varied between 0·95 and 1·05; in the last three weeks of December, a serie of twelve readings, among which there are a quite unusual number of lo readings and among these one significantly low reading; in the first fort night of January, again, nine readings, of which eight fall below th normal, one again being significantly below the normal; then, in the las week in January, a fluctuation which has all the characters of an inocula tion or auto-inoculation curve—i.e., a negative phase, a positive phase, and a secondary ebb; and, lastly, throughout the rest of the chart a very significant series of large oscillations in which very low readings are inter spersed with isolated high readings.

Chart 5 (*continued*).

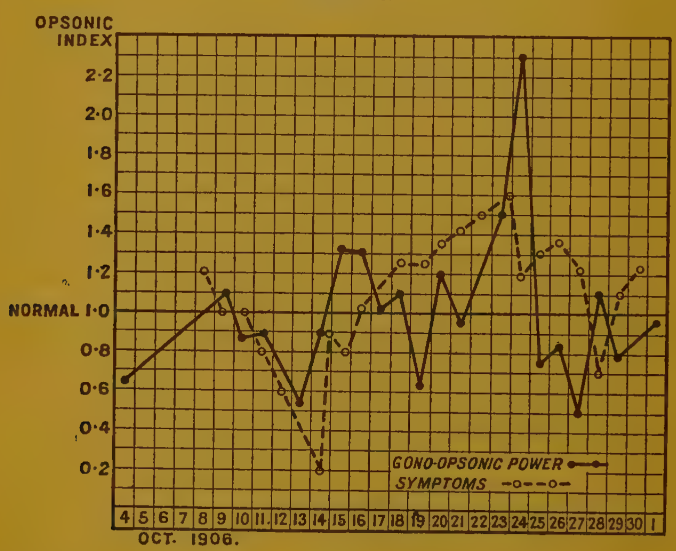


Of special import in connexion with this chart are the following fact
Early in the last week in January—i.e., in the week in which the fir
really significant fluctuation was registered—a lymphatic gland in th
submaxillary region began to swell and to give a little trouble. After
few days this trouble subsided and the gland could no longer be fel
Early in the second week of February one of the physicians made a caref
examination of our fellow-worker's chest. His examination gave onl
inconclusive results. About the date on which the chart here terminat
tubercle bacilli were detected in our fellow-worker's sputum.

III.—Chart showing, in connexion with a case of Gonococcal Arthriti
Auto-inoculation and Immunising Response, and the Correspondenc
which exists between these Phenomena and the Clinical Events.

Chart 6.—This chart furnishes a record of the measurements of th
gono-opsonic index which were made in connexion with a case of gonococc

CHART 6.



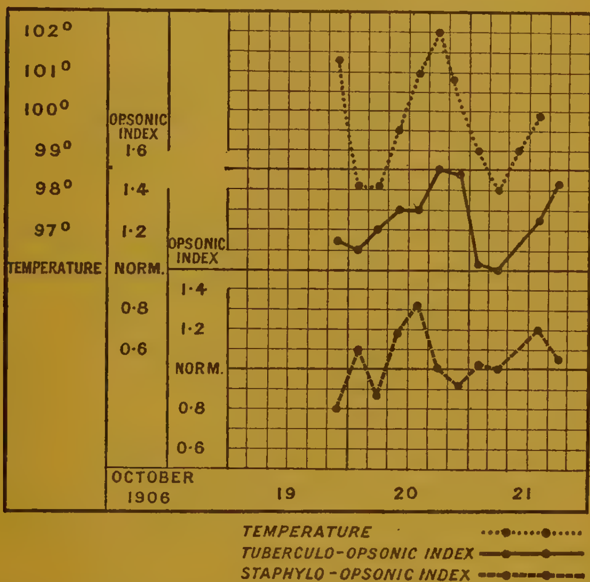
arthritis in St. Mary's Hospital. Along with the records of the bloo
testings we have entered upon the chart also a record of the daily stat
of the patient. The two records were obtained by different observer
working without any intercommunication. Dr. J. Freeman, who under
took the blood examinations, did not enter the wards, while Mr. K. Lees, wh
undertook the record of the symptoms, was engaged exclusively in clinica
work. The clinical record was obtained in the form of the curve her
charted by assigning a daily quota of marks to the patient in accordanc
with a scheme previously agreed upon. Very convincing, when considere

the light of these facts, is, we think, the testimony of the chart to the se interrelation between the content of the blood in protective ostances and the clinical state of the patient. (Compare here the ents that are put on record in connexion with Charts 22 and 23 d 33.)

.—Charts showing Auto-inoculation Effects in connexion with Advanced Phthisis.

Chart 7 has reference to a case of phthisis. The patient was in the t stage of the disease and was affected with a typical hectic pyrexia. s sputum, which had been frequently examined, was characterized by e association of numerous staphylococci with the tubercle bacillus. In

CHART 7.

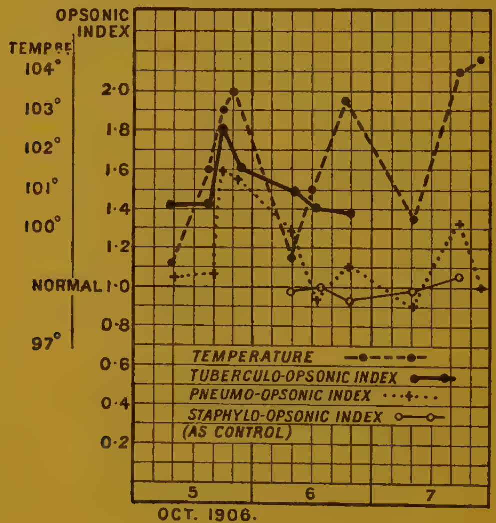


upper division of the chart we have the temperature curve ; in the ldle division the record of the observations which were made on his erculo-opsonic power ; and in the lowest division of the chart the record the changes in his staphylo-opsonic power. It will be seen that the and fall of the temperature here coincides with a rise and fall in the erculo-opsonic power, and that it corresponds also in a general way with uctuation in the staphylo-opsonic power.

Chart 8.—This chart has reference to another patient also in the last ge of phthisis. This patient's sputum had been continuously free from ohylococcus, while it contained on each occasion in addition to tubercle

bacilli innumerable pneumococci. It will be seen that while the tubercul
and pneumo-opsonic power of the patient's blood rose and fell with h
temperature, his staphylo-opsonic power remained unaffected. It
interesting to note that in an earlier period of his disease—when, howeve
the hectic temperature had already persisted for over six months—the
was no daily variation in his tuberculo-opsonic power, while his pneum
opsonic power rose and fell with the diurnal variation of the temperatur

CHART 8.

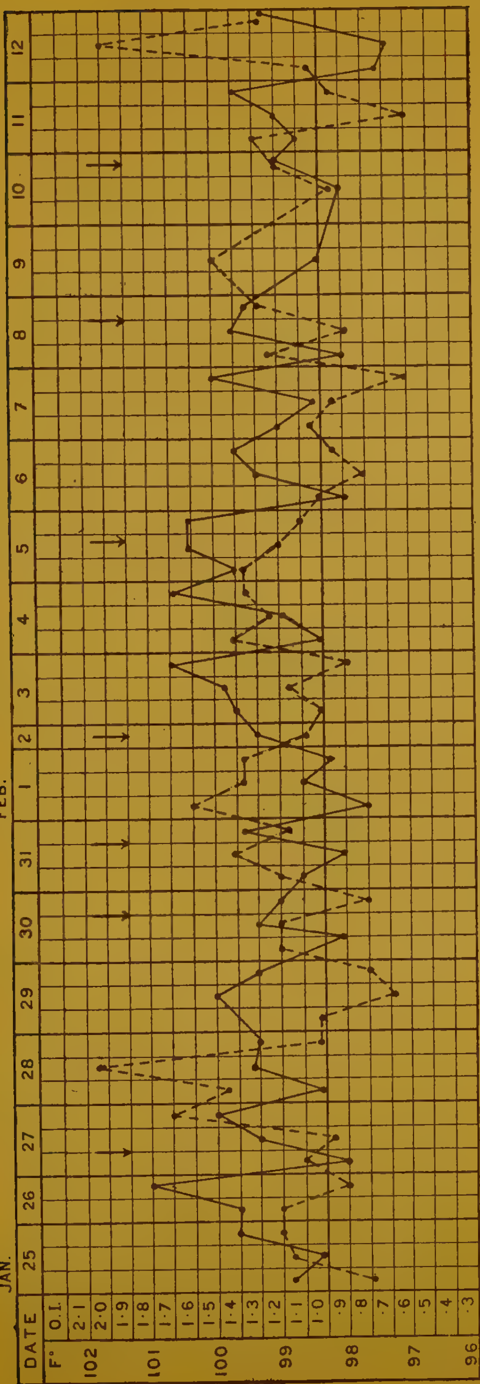


Charts 9 *and* 10.—These charts which we owe to Dr. A. C. Inman, w
worked with us for a long time and who is now in charge of the opson
department at the Brompton Hospital for Consumption, refer to two cas
of pulmonary phthisis which were treated with tuberculin under t
control of the opsonic index. These records are introduced here as sho
ing in a quite typical way that inverse relation of opsonic index
temperature which can be traced throughout the whole of the cha
which relate to septicaemic infection in man.

V.—Charts showing Auto-inoculation Effects in connexion with Gene alized Infections.

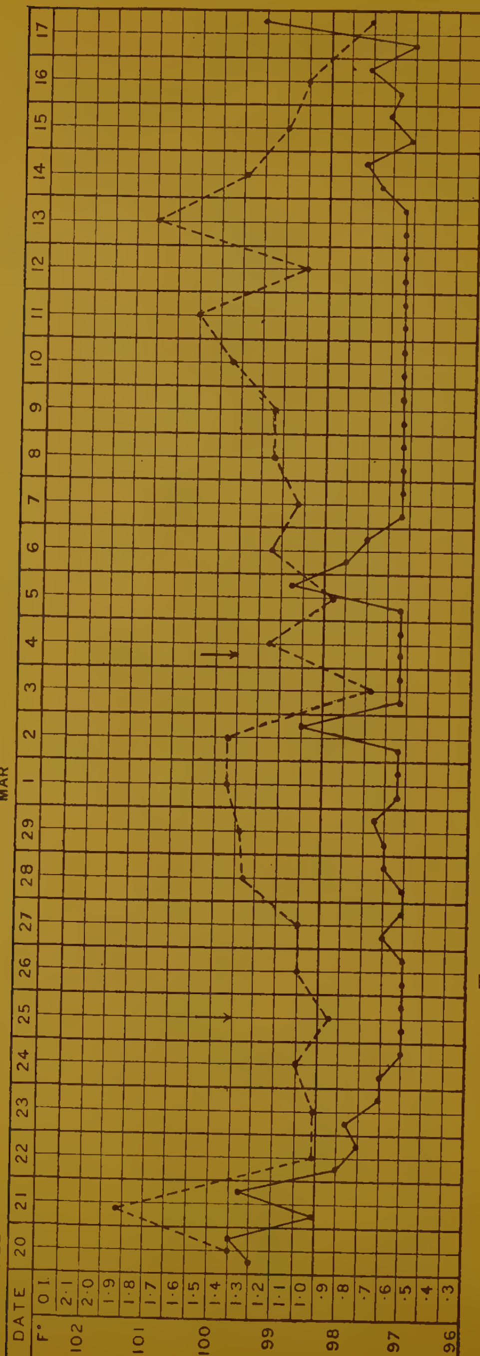
Charts 11 and 12 relate to two rabbits which were inoculated in ea
case with half of an agar culture of living anthrax bacilli. There a
registered on the chart in each case the fluctuations of the anthrac

CHART 9.



Temperature ——— Tuberculo-opsonic index - - - - -

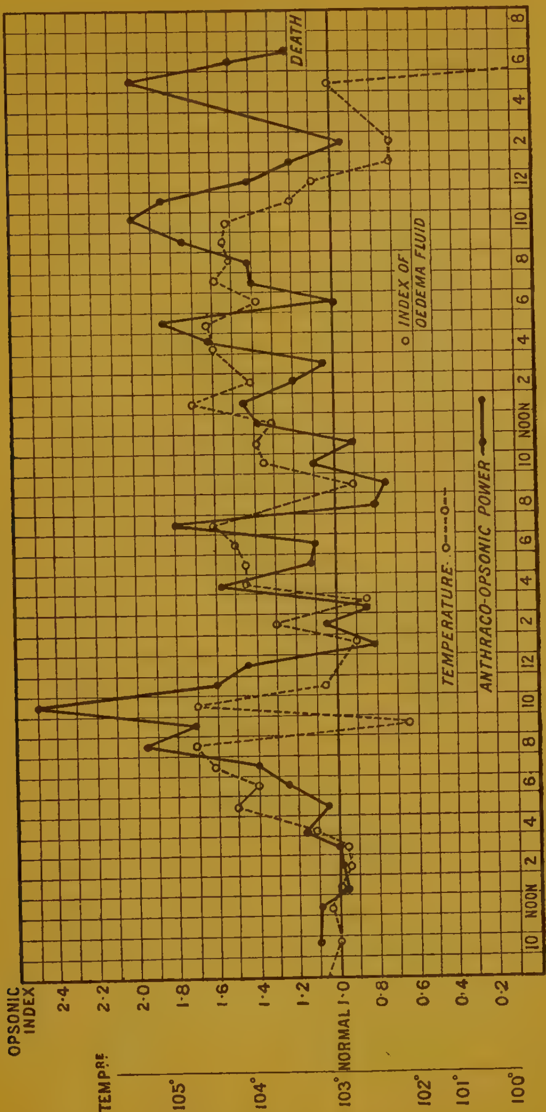
CHART 10.



Temperature ——— Tuberculo-opsonic index - - - - -

sonic power[1] of the blood in association with the temperature readings,

CHART 11.



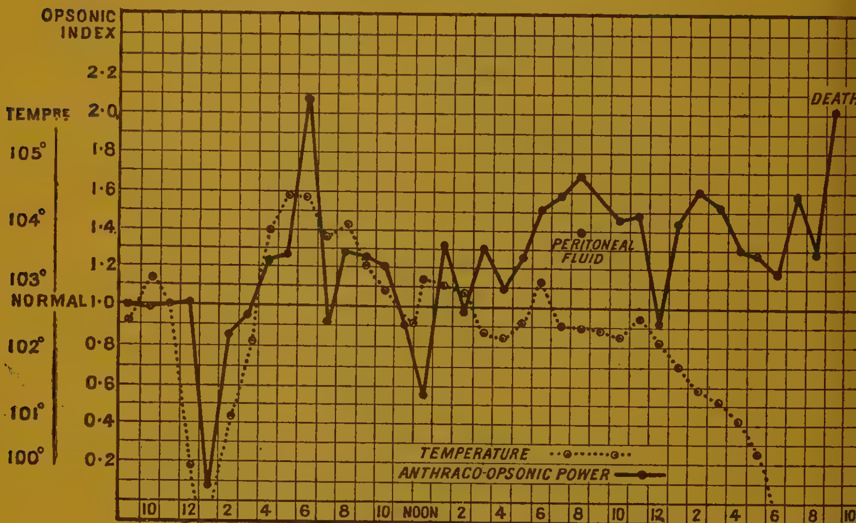
[1] The technique which was employed for the estimation of the anthraco-opsonic power of these rabbits' blood differed from the ordinary technique (*a*) in the respect that a suspension of spores was here substituted for the customary bacterial suspension; and (*b*) in the respect that the opsonic index was here arrived at not by comparison with the blood of a normal animal, but by comparison with blood drawn off from the rabbits before the outset of the experiment, or with a blood which had been standardized against this.

which were taken in the rectum. The more important points which are brought out in the charts are the following :—

1. The animal organism is shown to be capable of making immunising responses even to such acute infections as were here in question.

2. The fluctuations of the body temperature are shown to correspond here in an astonishing manner with the fluctuations of the opsonic power of the blood.[1]

CHART 12.



3. The generalization that the bacterio-tropic pressure in the focus of infection is normally lower than that in the circulating blood is here corroborated by the fact that the fluid drawn off from the site where the inoculation was made had in each case a smaller opsonic power than the circulating blood.

4. A comparison of Chart 11, where the machinery of immunisation shows sign of flagging at the close, with Chart 12, where the machinery of immunisation continues to respond in an active manner, shows that death in acute infectious disease is not always the result of a breakdown in the machinery of immunisation. It may also follow upon the breaking down of some other part of the physiological machinery. (*Vide* here Chart 17.)

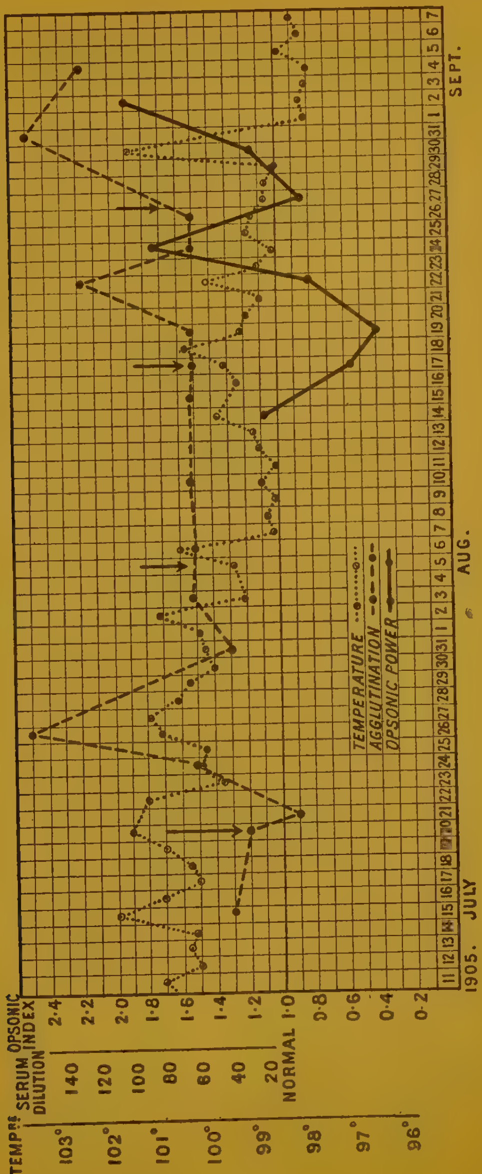
VI.—Charts showing (*a*) the Conditions which are met with in Man in Streptococcal and other Septicaemias, and (*b*) the Results which have been obtained in these Conditions by Recourse to Vaccine-therapy.

The subjoined group of charts depict a type of septicaemia which

[1] A quite similar correlation between temperature and immunising response is exhibited in Charts 7 and 8.

ould appear to differ fundamentally from the type depicted in Charts and 10. A study of so much of the Charts 13, 14, 15, 17, and 18 as

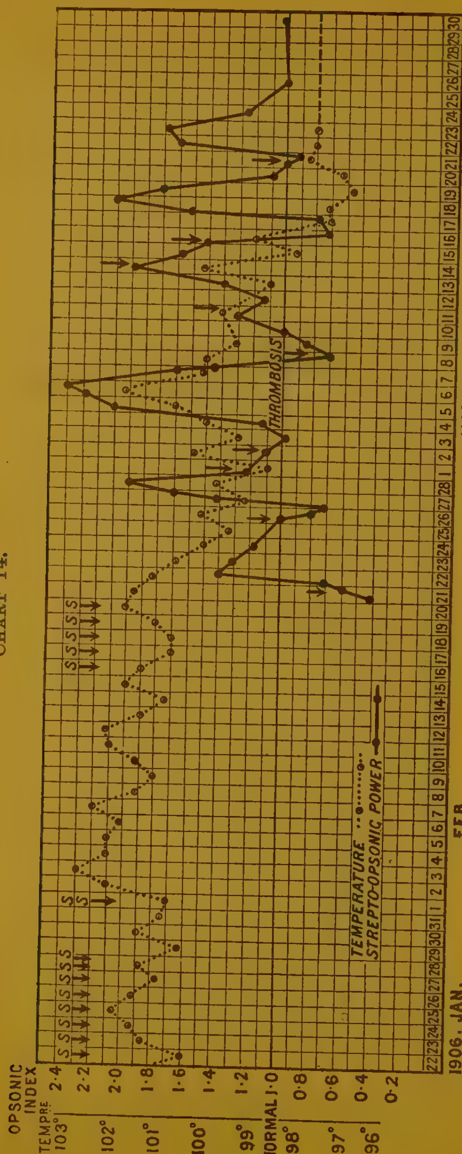
Chart 13.



refers in each case to the untreated condition of the patient, and of the whole of the Charts 16 and 19, shows that the spontaneous immunising

responses which are so salient a feature in connexion with Charts 7 and 8 and 11 and 12 here make default. In view of this fact—and it is a fact

CHART 14.



S with an arrow—injection of "anti-streptococcus" serum. Arrow alone—injection of streptococcus vaccine.

which assuredly must stand in association with the long duration of all these septicaemias and the fatal character of streptococcal septicaemia—we

made in each of the cases which are charted below a methodical attempt to evoke the required immunising responses by the subcutaneous inoculation of bacterial vaccines.

Chart 13 applies to a case of Malta fever which had been running the lingering course which is characteristic of so many cases of this disease. As is the rule in such lingering cases, the patient's serum was found to possess only a very moderate agglutinating power—agglutination effects being incompletely developed in all dilutions above 1 in 40. This being the situation of affairs, a series of inoculations of a vaccine made from a sterilized culture of the micrococcous Melitensis was undertaken, the effect of these being watched by blood examinations and temperature observations.[1]

In connexion with the first two inoculations the examination of the blood was confined to the measurement of the agglutinating power. In the case of the last two inoculations—when the difficulties in connexion with the opsonic technique as applied to the micrococcus Melitensis had been successfully overcome—examinations of the opsonic power were added. The chart shows that an increase of the bacteriotropic substances of the blood was obtained by vaccine-therapy, and that this went hand-in-hand with a decline of the temperature—this last remaining permanently normal after the date at which the chart ends.

Interesting also is the fact that, as might have been expected, the temperature rises in the negative phase when the bacterial substances which have been injected are no doubt circulating in the blood.

Finally, it is to be noted that there is a general agreement between the rise and fall of the agglutinating power and the rise and fall of the opsonic power.

Chart 14.—The details of the case to which this chart applies have already been published in *The Lancet*[2] by Sir James Barr and Dr. Blair Bell conjointly with one of ourselves. It will suffice in elucidation of the curve here published to rehearse the more interesting features of the case.

The patient, a young woman, was taken ill some three or four weeks before the date at which the chart commences with an acute sore-throat, accompanied by high fever. In association with the condition in the throat an acute swelling developed in the side of the neck. In connexion with this the question of incision was considered. Before this could be carried out the temperature, which had been very high and continuous, became lower and remittent, and the swelling rapidly disappeared. A cardiac murmur now developed and the case assumed all the characters of an infective endocarditis. Treatment by "antistreptococcic" serum was now resorted to by Sir James Barr and Dr. Blair Bell, and

[1] It is to be noted that the temperatures which are recorded in this and the subsequent charts are in each case average temperatures representing the mean of all the observations which were made in the course of the twenty-four hours.

[2] *The Lancet*, February 23, 1907, p. 499.

was persisted in daily on eight successive days, as indicated on the chart, without any lowering of the temperature. After several attempts to obtain a culture from the blood had proved abortive a

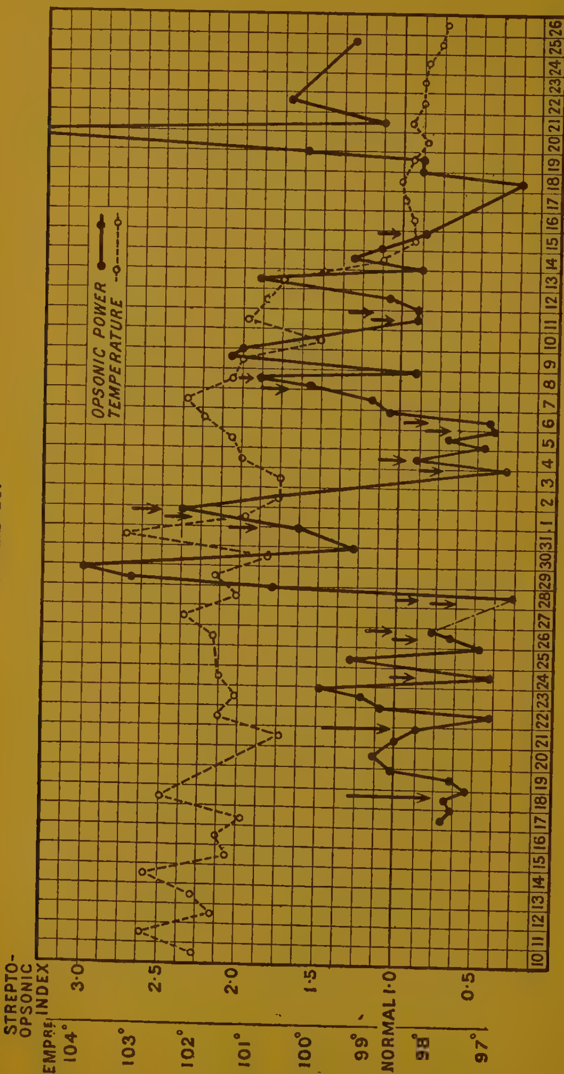


Chart 15.

culture of streptococcus was on February 12 obtained. Following upon this finding, "anti-streptococcic" serum was again resorted to by Sir James Barr and Dr. Blair Bell. On February 21, the day following the last of the second series of serum injections, two measurements

of the patient's opsonic power were made with a culture of the streptococcus derived from the blood of February 12. It will be noted that in spite of the fact that large quantities of "antistreptococcic" serum had been administered the patient's opsonic index proved to be very low. Vaccine-therapy was now resorted to, a vaccine made from cultures of the patient's streptococcus being employed in doses of from 5,000,000 to 12,500,000. The favourable result of this treatment can be appreciated on noting in the chart how the inoculations were followed by very striking immunising responses, and how in association with the increase in the opsonic power the temperature rapidly declined[1] until within little more than three weeks after the first inoculation it came down to, and remained permanently at, the normal level, the patient making an uninterrupted recovery without any subsequent relapse.

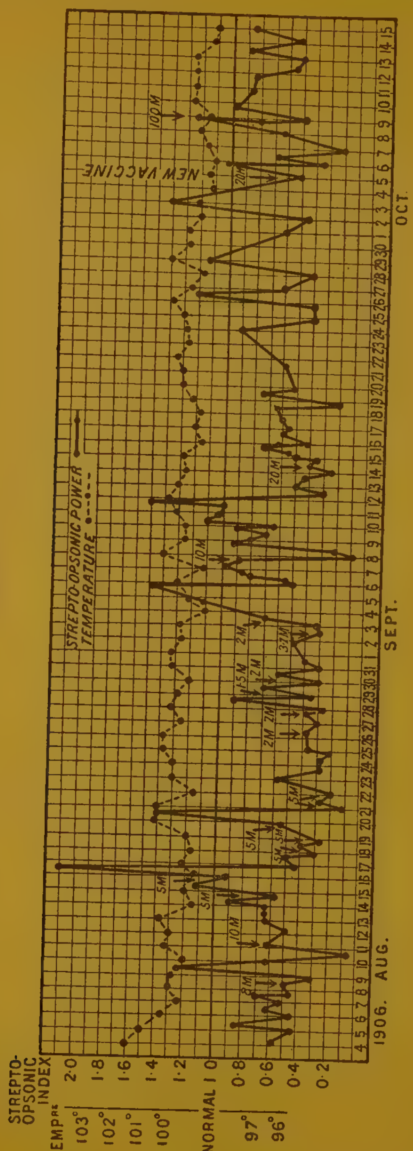
Chart 15.—The patient to whom Chart 15 applies was admitted to St. Mary's Hospital early in May, 1906, with phlegmonous abscesses and a high temperature. Soon after admission her condition became extremely critical, new abscesses developing in different parts of the body and her temperature ranging very high. Cultivations made from the blood remained sterile, but streptococci were recovered in pure culture from the abscesses. The patient's strepto-opsonic power as tested with these cultures ranging between 0·6 and 0·7, it was decided to see whether anything could be done for the patient by the inoculation of a vaccine which had been prepared from her streptococcus. The effects which were obtained by this means and the relation in which the patient's temperature stood to the opsonic power of the blood, can be followed upon the chart.

A study of the chart will suggest that the low opsonic power recorded on May 17 stood in relation with the high temperature of the 18th; that the rise of the opsonic power on the 20th stood in relation to the decline of the temperature on the 21st; that in a similar manner the low opsonic reading of the 22nd stood in relation to the rise of temperature that day; the rise of the opsonic power on the 29th to the lower temperature of the 30th; the low opsonic reading of the 31st to the higher temperature of June 1; the high opsonic reading of June 1 to the decline of temperature on the next two days; the low opsonic readings of the 4th, 5th, and 6th to the rise of temperature of those days; the high opsonic readings of the 8th and 9th, with the lower temperature of the 10th; again, the low opsonic readings of the 11th and 12th, with the high temperature of those days; and, lastly, the high readings of the 13th and 14th, with the defervescence which took place on those days.

[1] It is of incidental interest in connexion with this case that a thrombosis which developed in the left iliac vein in the course of the treatment, sending up the temperature in the manner shown in the chart, was very rapidly and effectually dissipated by the administration of large doses of citric acid, as recommended by one of us. ("A Note on the Causation and Treatment of Thrombosis occurring in Connexion with Typhoid Fever." (*The Lancet*, December 6, 1902, p. 1531.)

And there is more than this. The record of the new abscesses which developed after the patient came under our observation shows that

CHART 16.



these developed on May 17th, 18th, 20th, 26th, 31st, and June 13th respectively. It will be seen on consulting the chart that May 17th, 18th, 26th, and 31st were all days on which low readings were

obtained, and that May 20 and June 13 were both days which followed immediately upon days when low opsonic readings were obtained. It would on general grounds seem to us very probable that days when the citadel of the blood is not firmly held against bacterial invasion would be likely to be days in which bacteria could be conveyed unharmed from place to place by the channel of the circulation.

We have only to add with respect to this case that after the defervescence of June 13th and 14th the temperature never rose again, the patient leaving hospital early in July in a very satisfactory condition.

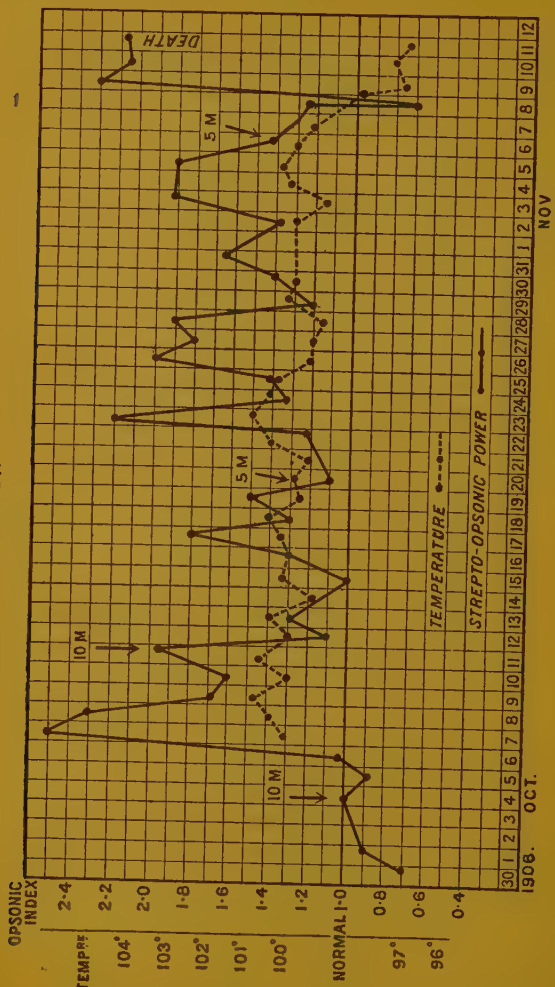
Chart 16 refers to a patient, aged about thirty-five years, who was admitted to St. Mary's Hospital on July 31, 1906, with symptoms of cerebral embolism, pyrexia, and systolic murmur. The history which was obtained was to the effect that while engaged in her ordinary work twelve weeks previously the patient had been suddenly seized with facial paralysis and complete hemianaesthesia. A culture of streptococcus having been obtained from the blood and nine successive measurements of the patient's strepto-opsonic index having shown that she was not making any spontaneous immunising response, vaccine-therapy was resorted to. The vaccine was, of course, made from the patient's streptococcus. In spite of the fact that an almost lavish amount of labour was expended upon this case ; in spite of the fact that we reached high and reached low to find an effective dose of our vaccine ; in spite of the fact that we essayed with small doses frequently repeated and with large doses spaced far apart ; in spite of the fact that we went over the ground a second time and made fresh cultures from the blood, recovering again a streptococcus, and prepared a new vaccine,—in spite of all this, and in spite of the fact that the patient was when we commenced relatively speaking strong and vigorous, all our efforts to elicit immunising responses were as good as unsuccessful, the opsonic power of the blood remaining almost continuously below the normal line. The clinical event was in conformity with this. The patient, becoming gradually weaker, went home and died within a week after the date upon which our chart terminates.

When we compare this case and the case to whom Chart 15 refers, and when we remember that that patient, though enfeebled and almost moribund, made vigorous immunising response and recovered, while this patient, though comparatively vigorous and strong, made no such response and succumbed, the importance of immunising response cannot but be very forcibly suggested to us. The lesson that immunising response is an all-important factor in recovery will be seen to be the lesson which is taught by all our cases.

Chart 17 refers to another case of streptococcal endocarditis. The patient, a man aged over fifty years, had at the date at which the chart begins already undergone three months' continuous fever associated with progressive aortic mischief and cardiac dilatation. A culture of streptococcus was obtained from his blood and vaccine-therapy was embarked upon. The chart shows that the patient immediately responded

with an increase in his opsonic power, but that for a whole month no effec
was produced upon his temperature. During that month the cardia
dilatation was making steady progress, and towards the end it becam
clear that the cardiac muscle was on the point of giving out. Finally, and
it would seem in association with the immunising response to the las

CHART 17.



noculation, the temperature became, first, normal, and, then, subnormal. At this point death stepped in and terminated the scene.

It would seem to us that the lesson of Chart 12—the lesson that death may supervene in bacterial infections independently of any breakdown in the machinery of immunisation—is here strikingly corroborated.

Chart 18 relates to a patient, aged twenty-five years, who was sent up to St. Mary's Hospital for inoculation treatment from Winchester Hospital.

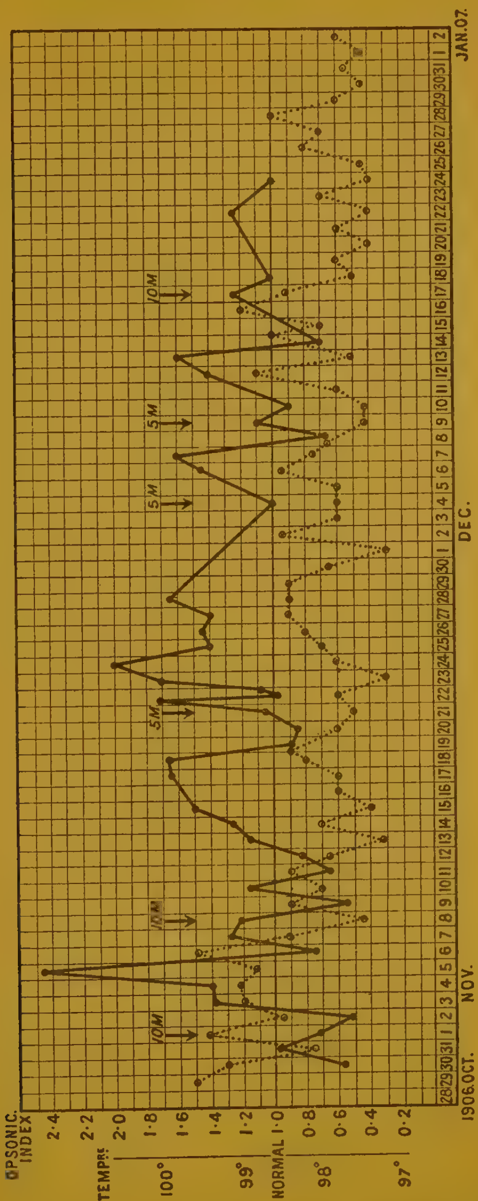
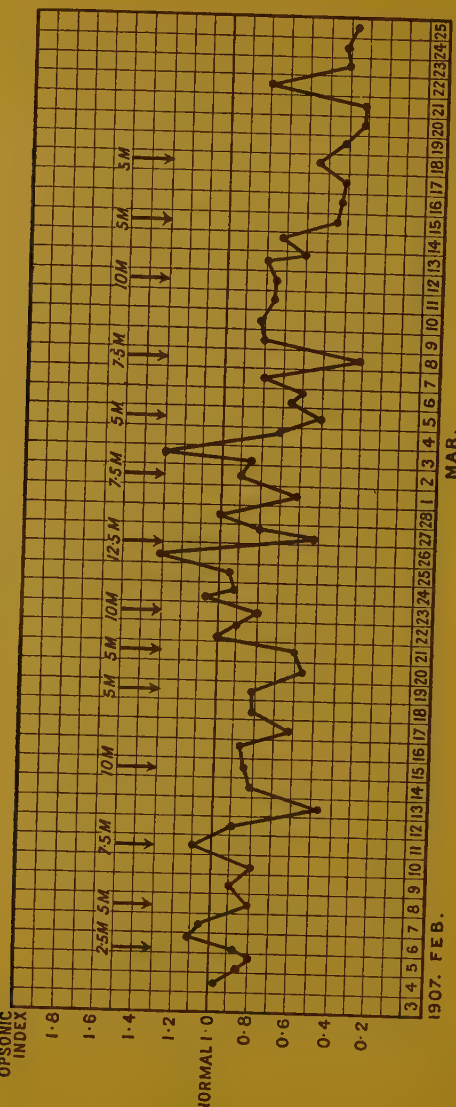


CHART 18.

A culture of staphylococcus had there been obtained from her blood. She had a history of eight months' occasional pyrexia and of pulmonary

infraction and cerebral embolism arising in connexion with obvious mit
disease. On September 27, 1906, the day after the patient had been a
mitted to St. Mary's Hospital, cultures were made from the blood, with

CHART 19.



negative result. None the less, in view of the bacteriological findings at
Winchester Hospital, and the fact that her index was fluctuating to
staphylococcus, it was thought well to inoculate with staphylococcic

accine. On October 17 she was injected with a small dose of the vaccine hich we were employing in the treatment of ordinary staphylococcal fections. On October 27, during an exacerbation of her pyrexia, cultures ere again made from the blood. These revealed the presence of a aphylococcus which possessed in several respects quite aberrant charcters. A vaccine having been prepared from this staphylococcus the oculations were resumed with the result that the patient's temperature on became normal and remained permanently normal after the date which the chart ends.

As will be seen on reference to the chart, the inoculations elicited in is case on the whole quite satisfactory immunising responses, the tient's staphylo-opsonic index maintaining itself almost constantly ove the normal line.

Chart 19.—The patient to whom this chart refers was a man aged thirty ars, who had many years before developed a mitral lesion after an tack of "rheumatic fever." Commencing insidiously with pyrexia d weakness the septicaemic attack which is here in question gradually unasked itself as ulcerative endocarditis, the cardiac symptoms developing st in connexion with the mitral valve and afterwards involving the rtic valves. After repeated unsuccessful attempts a small diplococcal reptococcus was cultivated from the blood early in February. A vaccine ving been prepared from this, inoculation treatment controlled by ily measurements of the opsonic index was embarked upon. It was soon cognized that the patient made no satisfactory immunising response small doses, and that larger doses produced constitutional disturbance sociated with pronounced negative phases. None the less, after me three weeks' treatment, a certain amount of improvement made elf manifest, and some slight hope was entertained for the patient. iese hopes, however, proved illusory, immunising response to inoculan was conspicuous by its absence, and a steady downward tendency ide itself manifest both in the opsonic power and in the patient's ndition. Vaccine-therapy was accordingly abandoned and soon after e date at which the chart terminates the patient succumbed.

It is again very clearly brought out by the history of this case that iere inoculation fails to evoke immunising response it is entirely useless.

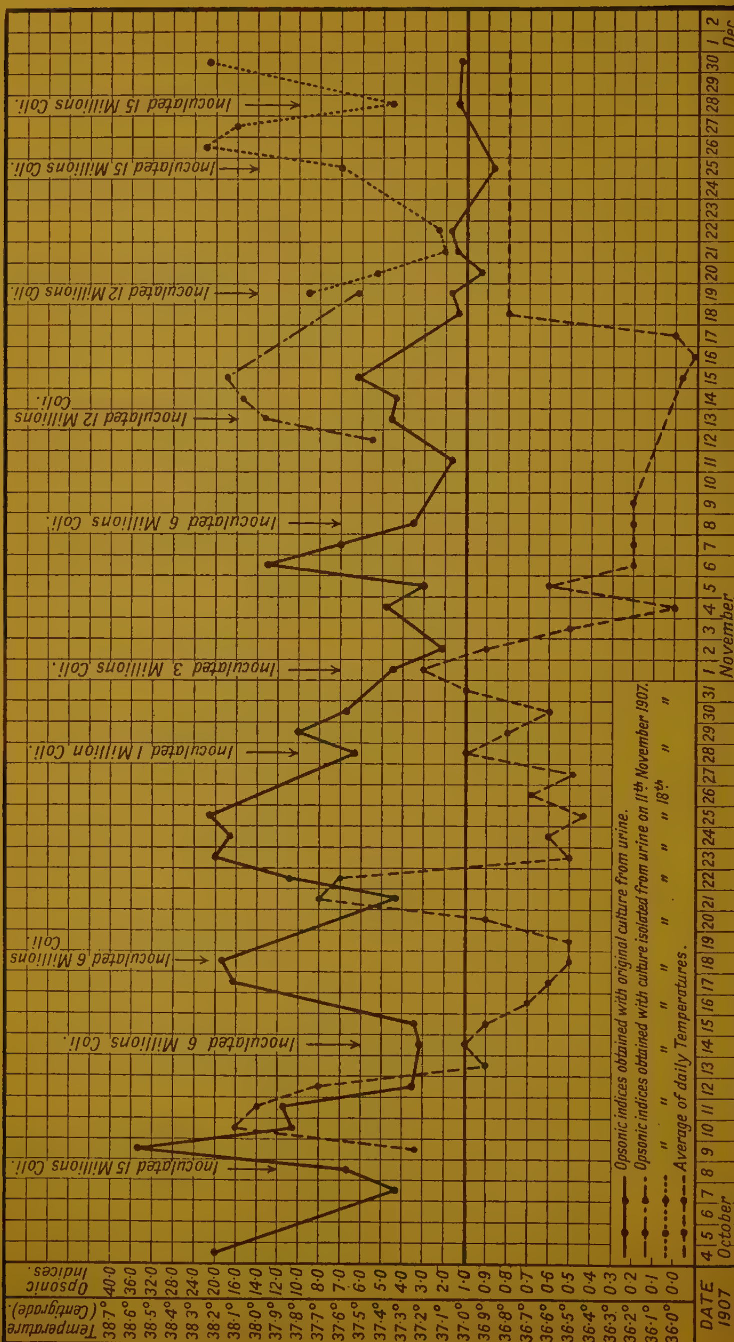
Chart 20 refers to an old gentleman who had been suffering for years om coli-cystitis and enlargement of the prostate, and who had long en unable to pass urine without recourse to the catheter.

For many months previous to the date at which the curve begins had suffered from attacks of fever recurring at intervals of one to o or three weeks. These were rapidly reducing his strength, and oduced great discomfort, owing to the necessity for very frequent ourse to the catheter.

He had long been treated unavailingly with urinary antiseptics d by daily washings of his bladder.

When proceeding to London for treatment by vaccine-therapy he

CHART 20.


Inoculated 15 Millions Coli.
Inoculated 6 Millions Coli.
Inoculated 6 Millions Coli.
Inoculated 1 Million Coli.
Inoculated 3 Millions Coli.
Inoculated 6 Millions Coli.
Inoculated 12 Millions Coli.
Inoculated 12 Millions Coli.
Inoculated 15 Millions Coli.
Inoculated 15 Millions Coli.
Opsonic indices obtained with original culture from urine.
Opsonic indices obtained with culture isolated from urine on 11th November 1907.
" " " " " " " " 18th "
Average of daily Temperatures.
Temperature (Centigrade).
Opsonic Indices.
DATE 1907
October
November
Dec.

as, while travelling, attacked with fever, but managed to complete his urney. On arrival a pure cultivation of the bacillus coli was obtained om his urine, and on the next day his opsonic power to this microbe as measured. As shown in the chart it came out at 20·0.

This high opsonic index may, we think, be ascribed to an auto-nmunisation elicited by the auto-inoculation which produced the rise f temperature.

In view of the high reading which had been obtained inoculation as postponed until it should be found that the effect of this immunising esponse had passed off. A commencement was then made with a ose of vaccine corresponding to 15 millions of the patient's coli bacillus. nasmuch as some constitutional disturbance and increased irritability f the bladder supervened upon this inoculation, a reduced dose of 6 iillions was employed for the next inoculation. When as a result of his dose the index had been raised to 36·0 and the temperature had ecome sub-normal, we endeavoured by a repetition of this dose to cumulate n the direction of the positive phase. Instead of this the index fell way, and in association with this the patient again developed bladder rritability and a pyrexial temperature. This last made way for a sub-iormal temperature as the positive phase of the immunising response upervened, the irritability of the bladder at the same time quieting lown.

With a view to avoiding everything in the nature of constitutional eaction the dose of vaccine was on the next occasion cut down to 1 nillion. This resulting in nothing more than a very brief rise in the psonic index and a correspondingly brief lowering of the temperature, ollowed by a gradual decline of the opsonic index, and a corre-ponding increase of temperature, a further dose of 3 millions was dministered on November 1. This produced a very distinct effect in owering the temperature, but the opsonic index did not rise beyond 13·0.

All this time very marked progress was being made, the irritability of the bladder had quite disappeared, the catheter was employed only four times in the twenty-four hours, the patient was intellectually and physically more alert and was putting on flesh, and was quite losing the worn and anæmic appearance which he had originally presented. Furthermore the urine, which had been turbid with microbes, showed in a very characteristic way the phenomenon of agglutination, the microbes agglomerating into clumps large enough to be visible to the naked eye, and these as they rapidly settled down to the bottom of the urine glass left the whole upper portion of the urine absolutely water-clear.

On November 8 a further dose of 6 millions was inoculated without producing any visible rise in the index. On November 13, a further dose of 12 millions was administered producing only a trifling rise in the index. On November 19 another dose of 12 millions was administered without apparent effect on the index, and again on November 25 and

28, doses of 15 millions were administered without any effect on th index being registered.

All this time, in spite of the fact that the index was coming dow lower and lower, and in spite of the fact that the inoculations were doin nothing to raise it, the patient was making wonderful progress, gainin strength daily.

It was obviously impossible to refrain from seeking an explanatio of these paradoxical findings. It occurred to us that the microbe w were working with might be changing in the course of artificial cultiva tion, and might be becoming less and less resistant to the opsonic effec of the normal control serum, and accordingly that the apparent sinking away of the opsonic indices might quite well be due not to a diminutio in the numerator but to an increase in the denominator in the fractio $\dfrac{\textit{phagocytic count of the patient's blood}}{\textit{phagocytic count of the normal blood}}$ which gives us the opsonic index

With a view to putting this hypothesis to the test we cultivated th bacillus coli afresh from the patient's urine on November 11 and agai on November 18, and while the patient remained under observatio measured his opsonic power daily with respect to the original microb and freshly isolated microbe, obtaining the results which are show on the chart.

We may note, in conclusion, that since the patient returned home h has periodically been injected with small doses of coli vaccine, and h is reported to have remained marvellously well in every respect.

Chart 21 refers to a girl aged 22, who when seen by her physician o December 12, 1907, was found to be suffering from dilatation of th heart and a loud systolic murmur. (Two and a half years previousl he had ascertained that she had a reduplicated second sound at th mitral area.) The patient stated that she had developed "influenza " in November, and that she had not been able to shake it off. A pro- visional diagnosis of "ulcerative endocarditis, probably of influenza origin," was arrived at.

The patient was sent to bed. It was then found that she was suffering from an intermittent pyrexia. During December there were days when the temperature reached 102° and 103° F., and then the temperature came down almost to the normal for several days in succession. Early in January, 1908, the condition became more serious, with excruciating and continuous pains in the back and legs which necessitated recourse to injections of morphia.

On January 11, after intense and long-continued pain at the inner

[1] With a view to avoiding a possible source of error from this cause, ex- periments are now in progress with a view to seeing whether one and the same for- malinized bacterial suspension may not be employed where, as here, a prolonged series of tests are made with a microbe which undergoes attenuation on artificial media.

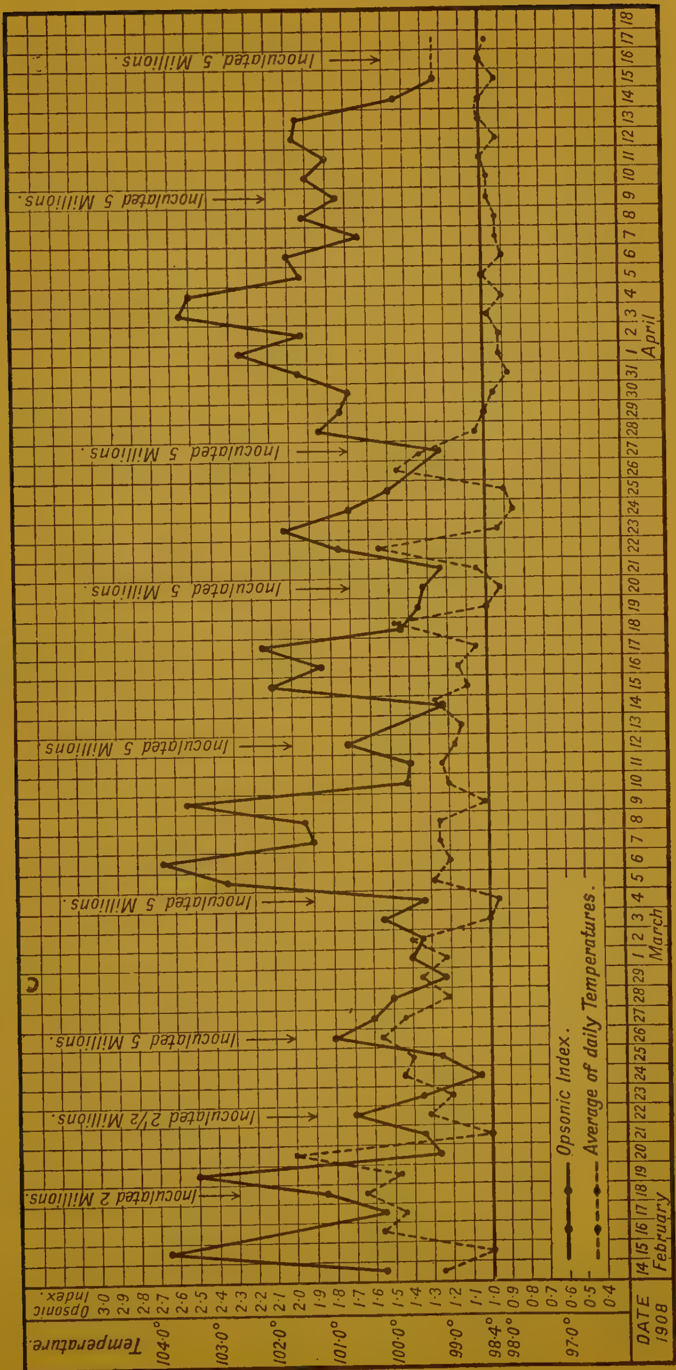

Temperature.
Opsonic Index.
Inoculated 2 Millions.
Inoculated 2 1/2 Millions.
Inoculated 5 Millions.
Inoculated 5 Millions.
Inoculated 5 Millions.
Inoculated 5 Millions.
Inoculated 5 Millions.
Inoculated 5 Millions.
Inoculated 5 Millions.
Opsonic Index.
Average of daily Temperatures.
DATE 1908
February
March
April

CHART 21.

side of the right ankle, an abscess developed here. The pus was ser for bacteriological examination and a negative report was received.

On January 12 there was a distinct rigor and the temperature ra up to 105° F. On this occasion, as on a previous occasion, in Decembe when the temperature ran up to 103° blood was drawn off from a vei in the arm by her medical attendant. On both these occasions th blood cultures were reported as sterile.

The patient's condition had now become extremely critical. Ther were frequent rigors and profuse sweating, a very dilated heart, a rapi pulse and a prolonged systolic murmur. Recourse was now had to hypo dermic and rectal injections of "anti-streptococcus" serum. These wer continued for ten days without any sensible effect on the temperature.

On January 18, after intense pain which had continued for days, small abscess developed on the left shin. Pus from this was sent fo bacteriological examination, and a report was received to the effect tha "a micro-organism resembling a pneumococcus" had been isolated, bu that the cultures had died out.

In consequence of this report the "antistreptococcus" serum injec tions were abandoned, and injections of "antipneumococcus" serum anc a pneumococcus vaccine were resorted to. On January 19, after sever headache which had continued for days, an abscess about two centi metres in diameter developed on the forehead at the margin of the hair This turned black and sloughed, leaving a clean cut ulcer which healec very imperfectly.

On February 6 a new abscess developed on the right hand over th fifth metacarpal bone. A specimen of the discharge from this abscess and a few days later another sample of the same, were sent to one of us (A. E. W.) for examination. The specimen consisted in each case of a blood-stained transparent gelatinous fluid which showed under the microscope no micro-organisms and no formed elements, except here and there the remnants of a broken down leucocyte.

Cultures which were made from the first specimen yielded only ar isolated colony, in the case of the second sample only a very scanty growth. In the course of the study of this micro-organism it occurred to one of us (J. H. W.) that we might be dealing with the bacillus o glanders. This was afterwards confirmed when a brown growth was obtained on potato, and when a typical inflammation of the tunica vaginalis was produced in a male guinea-pig by intraperitoneal inocula- tion of the culture.

The fact that the patient's blood, a sample of which had been obtained for the purposes of the test, gave an opsonic reading of 1·6 rising next day to 2·65 made it practically certain that the micro-organism that had been isolated was the infective micro-organism which we were seeking.

On February 14 one of us (A. E. W.) went down to the country to see the patient. She was found in a very feeble and extremely emaciated condition, with a very loud presystolic murmur and an apex beat well

tside the nipple line. The wounds in the ankle and hand were found have healed by first intention, but there was some diffuse oedema the ulnar border of the dorsum of hand, such as might have suggested bercular disease. The wound on the shin and the ulcer upon the rehead were imperfectly scabbed over, and below the latter were a uple of indurated nodules, which, were it not for the other features of e case, might have been taken for acne indurata. Inquiry was now ade into the possibility of infection from horses, and it was elicited at the patient's pony had had "an influenza-like cold with a running the nose." The pony, which was one of three, was inspected, and as found to have a thin nasal discharge, and it was arranged that all e three ponies should be tested with mallein. Of the three one gave definite mallein reaction, and was shot, and was on post-mortem examination found to be suffering from glanders. The suspected pony did ot on this occasion give the mallein reaction, but subsequently eveloped definite signs of glanders, and was then shot.

A diagnosis of glanders having been established, a vaccine was made om the culture which had been isolated from the patient, and vaccine-herapy was begun on February 18, the opsonic power of the blood being easured daily. The details of the inoculations and of the malleo-psonic readings are shown upon the chart in association with the temperature curve which was obtained by taking the mean of the six daily hermometrical readings.

On March 3 the temperature, which had during the two previous nonths only rarely touched the normal, remained at the normal for a whole day. Since March 28 the evening temperature has not risen above 99 F. and since April 1 the patient's progress has been very rapid, the heart rate coming down to normal, the dilatation disappearing, and the patient putting on flesh rapidly, finally leaving for the seaside on May 11 to all appearance perfectly well.

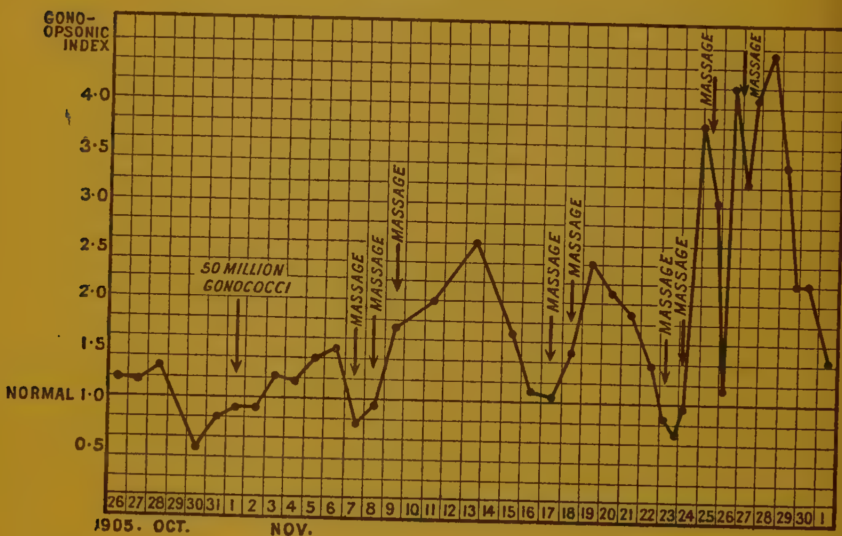
While recovery seems complete, there is reason to think that the glanders bacillus may not yet have been completely extirpated. Since the date on which vaccine-therapy was begun there have been the following evidences of the continued presence of the microbe in the system. On February 29 two small abscesses developed in the right axilla. After incision these healed by first intention. On April 3 the scar on the forehead broke down, and an abscess which had formed under it was incised and evacuated. On April 23 signs of activity manifested themselves in the scar on the left shin, and on May 9 a small amount of the same blood-stained gelatinous fluid as had been obtained from the other abscesses was evacuated. The observations of the malleo-opsonic power of the blood are still being continued.[1]

[1] At date of going to press (Oct. 1st, 1908), the patient is in perfect health. For the clinical notes relating to this case we are indebted to Dr. Hathaway, who was throughout in charge of the case, and who proposes later to publish a full account of the case in conjunction with one of us (J. H. W.).

VII.—Charts illustrating the fact that Auto-inoculation Effects are obtained where Active or Passive Movements are undertaken in connexion with Localized Bacterial Infections.

Chart 22 brings before the eye the result of the experiment which initiated our study of *induced auto-inoculations.* The chart relates to a case of gonococcal polyarthritis which was admitted to St. Mary's Hospital towards the end of October, 1905. As appears on the chart, vaccine-therapy was here embarked upon, a dose of gonococcal vaccine corresponding to 50,000,000 gonococci being administered on November 1.

CHART 22.



Six days afterwards, when it was proposed to administer a second dose of vaccine, it was found that the patient's knee had been massaged on the previous evening. In connexion with this the patient complained that while the massage itself had not been painful he had suffered six hours afterwards from constitutional reaction and from an aggravation of all his joint troubles quite similar to that which he had experienced a few hours after the gonococcal vaccine had been inoculated. The massage had, he said, "played him up cruel." Dr. Freeman, immediately recognizing that a negative phase effect due to massage might here be in question, thereupon desisted from his intended injection of vaccine, and took steps to investigate in a sytematic manner the effects of massage. The chart very plainly tells the history of his experiment. It is of fundamental significance to note, in the first place, that in association with the ample immunising responses which are recorded upon the chart the condition of the patient's joints improved with rapid

ides, and, in the second place, that although massage was confined the knees and ankles such joints in the upper extremities as were ected improved *pari passu* with the joints in the lower extremities.

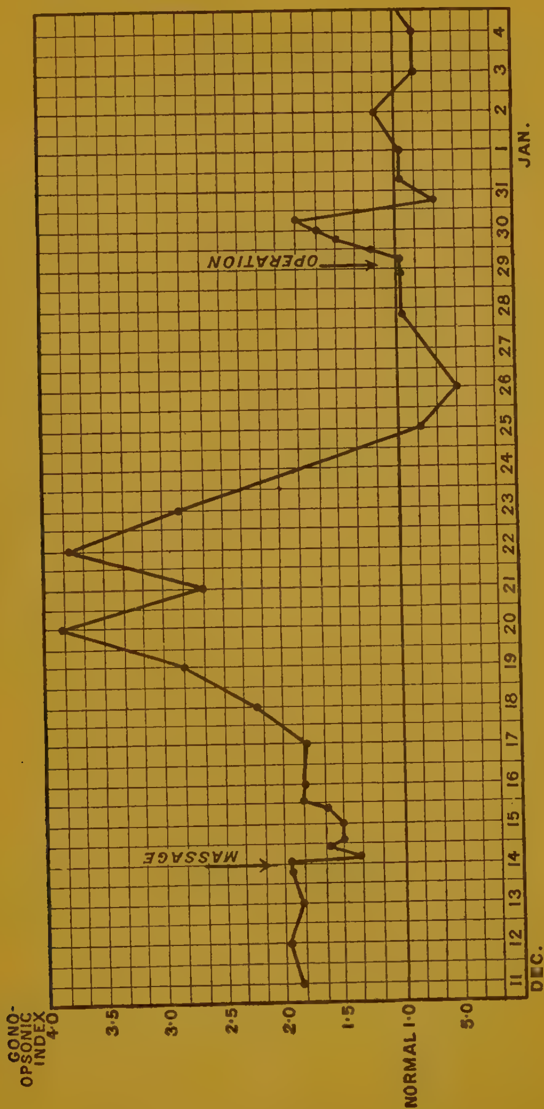


CHART 23.

The inference that this last result was attributable to therapeutic mmunisation is borne out by the fact that quite similar effects on emote regions of the body come under observation in connexion with nother method of therapeutic immunisation, to wit, with Bier's method

of passive congestion. (*Vide* in connexion with this commentary to Chart 43.)

Chart 23 has reference to a case admitted to St. Mary's Hospital for recurrent arthritis of the right knee which had supervened upon a gonorrhoea contracted some twelve years previously. The patient's gono-opsonic index was tested on five occasions, readings of 1·8 to 2 being on these occasions obtained. Although this of itself afforded strong presumptive proof of gonococcal infection, it was thought desirable before going further to make the diagnosis certain. With this view massage was prescribed. The operator who was employed was an enthusiast. After he had emptied the knee by his manipulations he expressed himself as satisfied that he had effected a cure. Fourteen hours later the left knee, which during the previous six months had been giving no trouble, began to swell and to become painful. This supervened in association with a fall in the gono-opsonic index and, possibly, as a consequence of a dissemination of living gonococci by the channel of the blood. Within twenty-four hours the left ankle also began to be painful, and, during the three subsequent days, the swelling increased in both joints; then, coincidently with the rapid rise in the opsonic curve which is shown in the chart, the swelling in both joints rapidly decreased. On December 29, when both knees were quite free from effusion, the right knee was opened up with a view to the removal of synovial fringes which interfered with the movements of the joint. An auto-inoculation effect, similar to those which will be considered in a subsequent section of this paper, followed upon this operation.

CHART 24.

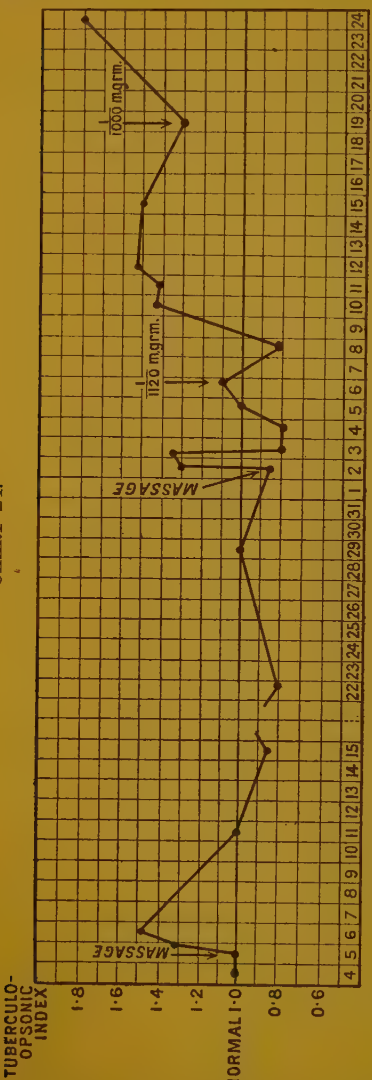
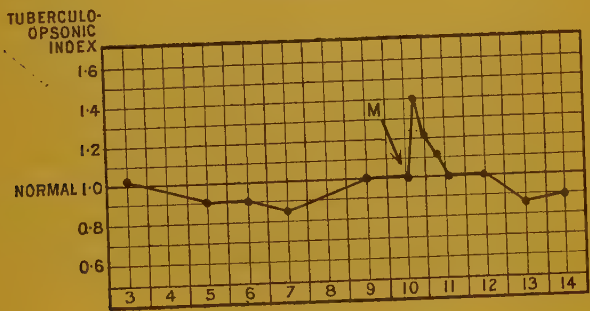


Chart 24 has reference to a patient, aged twelve years, who was admitted to St. Mary's Hospital with an arthritis of the knee which was

suspected to be of tuberculous origin. After the patient's tuberculo-opsonic power had been measured on two successive days, and had been found to be on each occasion normal, gentle massage was applied to the knee for half an hour. The result was to raise the boy's tuberculo-opsonic power from 1 to 1·5. About one month later, the knee having in the meanwhile been in plaster, gentle massage was again applied, with a somewhat similar result. Treatment by tubercle vaccine was now embarked upon. The effect which was produced upon the patient's blood by the first two inoculations is shown in the chart.

Chart 25 has reference to a patient who came to us for treatment with enormous masses of glands on both sides of the neck, and, further, glands in both groins. The patient had previously undergone six successive extirpation operations at the Middlesex Hospital. On one of these occasions the glands were microscopically examined and were pronounced to be tuberculous. After tuberculin treatment had been carried out by us for many months without appreciable improvement, it was resolved

CHART 25.

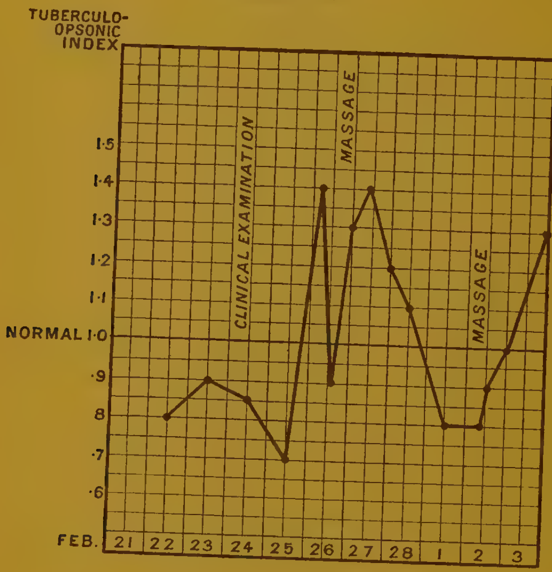


to test the diagnosis by massaging the glands. With a view to this, after an interval of weeks, during which all treatment was suspended, the patient's tuberculo-opsonic power was measured on six successive occasions, with the results shown in the chart. The glands in the neck were then gently massaged. Three hours after this the patient's opsonic index had risen from 1 to 1·4, falling then by stages to 1·3, 1·22, 1·12, and, finally, twenty-four hours after the massage to 1 as at the outset. Encouraged by the positive result of this diagnostic experiment vaccine-therapy with tuberculin was persisted in, with the result that a phenomenal improvement has now taken place in the patient's condition.

Chart 26.—The case to which Chart 26 applies was that of a young woman who came to us for treatment with enormous masses of glands entirely obliterating the outline of the jaw and chin. The patient was admitted to St. Mary's Hospital with a view to determining whether we had here to deal with tuberculous infection. In accordance with our programme, the patient was, previously to massage, to remain in bed

undisturbed for four days, her tuberculo-opsonic index being examine daily. This programme was followed out for three days, blood bein drawn off for testing on each morning. It, however, so happened o the afternoon of the third day that the visiting physician selected thi patient for a practical clinical demonstration, and that he called upo each member of his class to palpate the patient's glands. The effect o this palpation stands out clearly to view in the chart in the negativ

Chart 26.

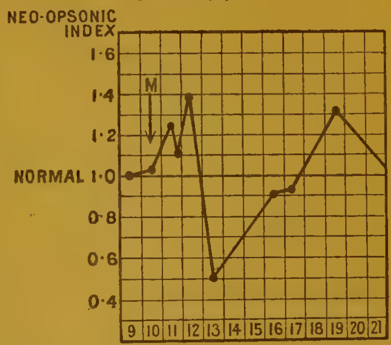


phase of February 25 and the positive phase of the morning of February 26. Quite unconscious of what had happened our part of the programme was followed out, the patient's blood being drawn off for examination on February 25 and again on the morning of February 26. Immediately after this latter sample of blood had been drawn we proceeded to carry out massage. The effect produced by this massage is seen in the chart in the low reading of the afternoon blood of February 26 and the positive phase and the secondary ebb which succeeded it. On March 2, when the blood had returned to its normal equilibrium, massage was again undertaken. In association with this there was again a marked fluctuation in the tuberculo-opsonic index.

Chart 27 relates to one of seventy or more cancer and sarcoma patients whom we have, with a view to the possible relief of some of their symptoms, treated with a vaccine made from that variety of staphylococcus which Doyen has brought into notice under the name of the *micrococcus neoformans*, attributing to it an etiological significance in connexion with tumour

ormation. The patient here in question was a woman, aged thirty-four ears, who was the subject of an inoperable sarcoma of the upper jaw which rojected through the unbroken skin of the cheek in the form of an orange. Vith a view to determining whether in this case anything could be hoped or from inoculations with a neoformans vaccine—and obviously such dvantage cannot be hoped for unless Doyen's microbe is present in he tumour—we here had recourse o massage and obtained the result which is exhibited in the curve. We ould here insist, in passing, that here appears to be very little room o doubt, in view not only of experiments like the above, but also in iew of the fact that the neo-opsonic ndex of cancer patients is continually fluctuating, that the so-called nicrococcus neoformans finds means o establish itself in a large majority f malignant tumours—the tumour forming in such case, no doubt nerely a *locus minimae resistentiae* to invasion by a microbe which nay be constantly present on the surface of the body.

CHART 27.



(For further examples of the induction of auto-inoculation by massage nd passive movements see Table under Serial Numbers 6, 7, 8, 9, 10, 13, 4, 15, 16, 18, 19, 20, 21, 22, 25, 26, 27 and 41.)

CHART 28.

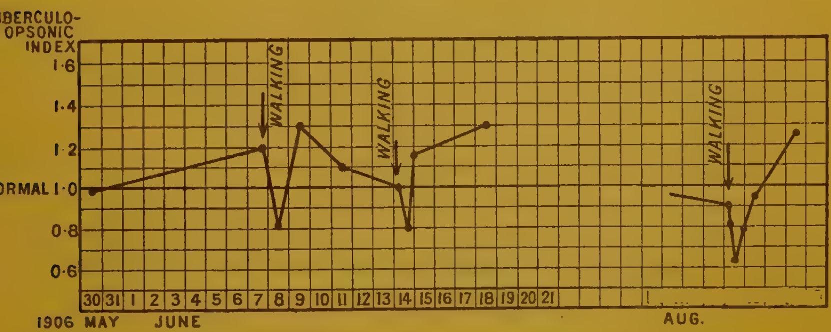


Chart 28 relates to a youth who presented himself for treatment for a mall sinus under the inner ankle of the right foot which refused to heal. The patient recounted that four and a half years previously he had pierced is foot with a sharp spicule of bone when he was digging into the ground cartload of bones obtained from the butcher as manure. The puncture thus made healed up and had broken open again. When this equence of healing over and breaking open again had repeated itself

several times the patient sought advice at St. Thomas's and afterwards St. Bartholomew's Hospital. He underwent in these hospitals three su cessive scraping operations. The foot had also been put up in plast and had been kept at rest upon a peg leg. The patient stated that had not had his foot to the ground for eighteen months. The patien tuberculo-opsonic index worked out as 1. He was instructed to return ten days' time in order that his tuberculo-opsonic index might be test before and after walking on the foot. We hoped by this means to lea whether the presumably tuberculous focus of infection in his foot was e tinct or aglow. Owing to a want of precision in our instructions the patie anticipated our wishes and discarded his peg leg when he had con within a short distance of the hospital and came hobbling into the labor tory. After withdrawing a sample of blood we set him to pace the hospit corridor for a matter of twenty minutes or more. Another sample of bloo was then withdrawn, and the patient resumed his peg leg. The readin which we obtained with these samples and with those obtained the ne day and again two days after will be seen in the chart. They furnished us presumptive evidence of the presence of tubercle in the foot. With view to further testing the diagnosis the peg leg, which had in the mea time been resumed, was on June 14 again discarded and the patient w again set to pace the hospital corridor for twenty minutes. The results the testings of the samples of blood withdrawn immediately before wal ing, immediately after walking, and—we think [1]—two to three hours aft walking, and again three days later, are displayed in the chart. We see the results confirmation of the diagnosis of tubercular infection in the foo Our diagnosis thus made, vaccine-therapy with tuberculin was begun. S weeks after, when the sinus had to all appearance completely healed, t peg leg was again discarded and the patient was set to walk the hospit corridor again for forty-five minutes. Samples of blood were drawn off f examination immediately before walking, half an hour, two hours, fo and three-quarter hours, ten hours, and again twenty-one hours aft walking. The results as set forth in the chart admonished us to contin our inoculations. After the treatment had been continued for seve months and again six months later, further tests of the patient's progre were made. The tuberculo-opsonic readings which were obtained c these occasions will be found chronicled in the Table, under Serial Nun bers 23 and 24. On both occasions the patient, who had long ago di carded his peg leg, undertook for the purpose of the test very activ exercise (quick walking and cycling). The inference arrived at aft the last testing—the inference, to wit, that the tubercle focus in the le was extinct—is apparently confirmed by the fact that the foot ha now for months been quite well—the last, and we hope final, incident i its pathological history having been the working out of a sequestru through the skin without a trace of suppuration.

[1] Our record here fails us.

Chart 29 relates to a youth with a sinus in the heel which was dis-
iarging through three minute openings. The successive tuberculo-
psonic readings which are recorded on the
iart on August 19 correspond respectively
samples of blood drawn off immediately
efore walking, immediately after walking,
id one hour after walking. There was, in
ldition to the readings that appear on the
art, also another reading on August 20
iich gave a value of 1·1.

CHART 29.

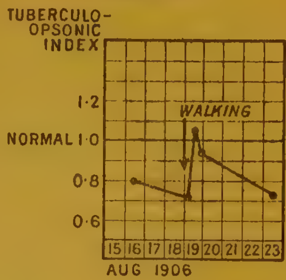
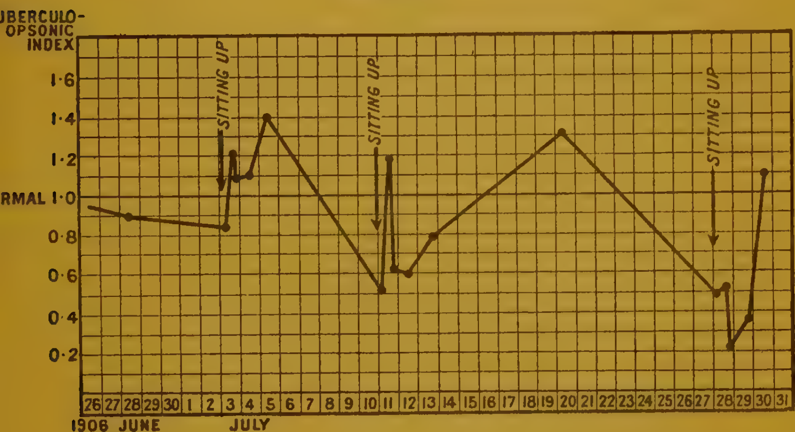


Chart 30 applies to a young medical man
th caries of the spine. After very marked
nprovement in his condition had been
hieved by vaccine-therapy the question
ose as to whether improvement had gone far enough to allow
his sitting up in bed. The patient was instructed to make
e experiment on July 3, withdrawing samples of blood for exami-
tion immediately before sitting up, half an hour after sitting up,

CHART 30.



ain six hours after, and then on the two following days. Ten days
erwards the experiment was repeated, and it was again repeated after
other interval of eighteen days. The results[1] of these tests seeming to us
warrant the inference that the focus of tubercle in the spine was still
low, we suggested to the patient that it might quite well be prudent,

[1] It will be observed that we have here registered both on July 3 and July 11
initial rise very similar to that to which attention was drawn in connexion with
art 1; and we have perhaps in the low initial readings of July 10 and July 18 in each
e a secondary ebb such as was illustrated in Charts 1 and 3.

E E

with a view to the avoidance of excessive auto-inoculations, to sit up or at intervals of ten days.

The next two charts disclose the fact that auto-inoculation effe can in cases of pulmonary infection be elicited by respiratory exertions.

CHART 31.

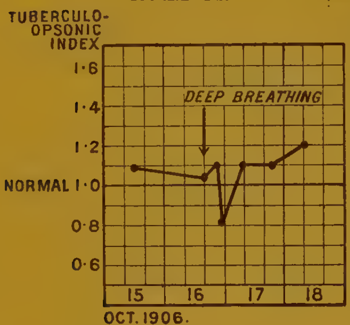


Chart 31 shows the fluctuation the tuberculo-opsonic index of a co paratively quiescent case of phthi which was elicited by a series of fo maximal inspirations and expiratio

Chart 32 shows the fluctuatio which were obtained in another case phthisis by a thorough clinical exar nation of the chest. The patient w in bed at the time of clinical exami tion and remained in bed during t period of observation. Immediat before the examination his tubercu opsonic index—and we may no dou ascribe this to an antecedent spontaneous auto-inoculation—stood 1·4, while his staphylo-opsonic index stood at 1. The chart clearly sho that there supervened here upon the deep breathing, coughing, a other respiratory exercises which are associated with auscultation characteristic negative and positive phase such as is obtained

CHART 32.

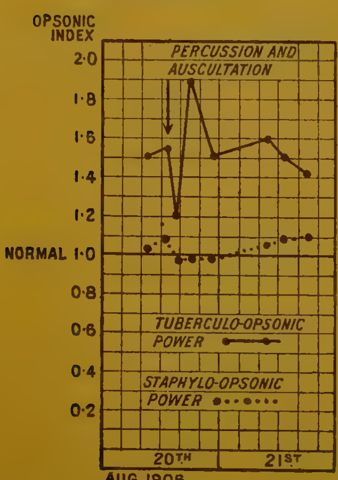
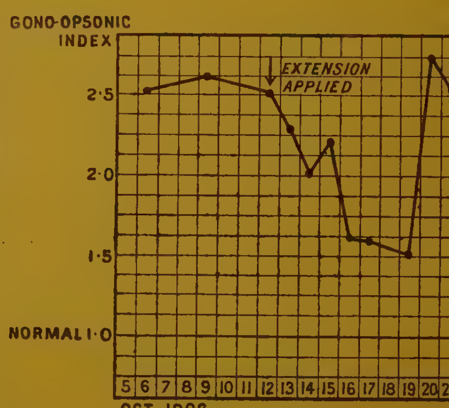


CHART 33.



the inoculation of tuberculin. The patient's staphylo-opsonic ind remained, as the chart testifies, throughout unaltered.

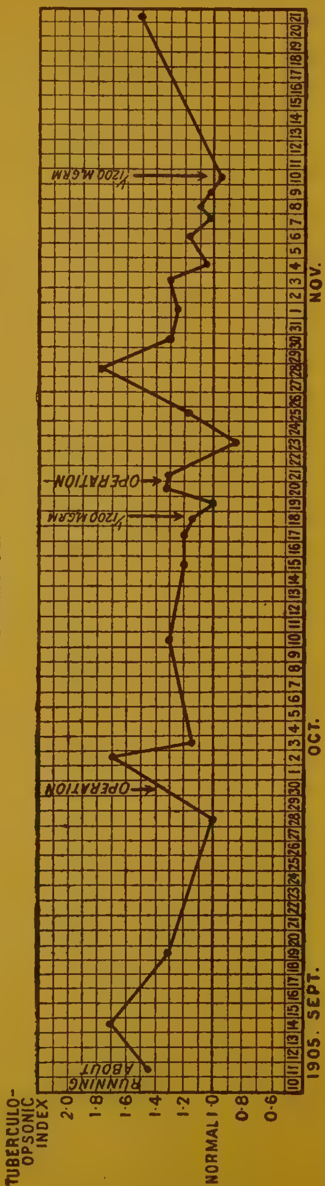
We think that it is of interest to note in connexion with the eviden which is here furnished with respect to the induction of an auto-inoculati

clinical examination of the chest in phthisis, that the results here ained are in harmony with the observation that periodical chest minations may in the case of phthisical ients be followed by constitutional turbance.

(For further examples of the induction uto-inoculations by active movements infected parts see Table under Serial mbers 1, 2, 3, 4, 5, 23, 24, 30, 34 and 35.)

Chart 33 refers to a patient who was nitted to St. Mary's Hospital on Octo- 6, 1906, with gonococcal arthritis, the ee being rigid and flexed. The ient's gono - opsonic index having n measured on October 6 with the ult shown upon the chart, a dresser s charged to apply extension. The ient's gono-opsonic index was again asured on the 9th and 12th. As the rt shows, practically the same values e obtained as on the first occasion. October 12 the surgeon in charge of case examined the apparatus, found t it was not exerting any pull upon leg, and proceeded to make the ension effective. Upon this the patient an to complain of general malaise and pain in other joints. In association h this we registered the fall in the o-opsonic index which is shown on the rt. External circumstances made it ossible to follow the curve beyond the nt at which it here terminates.

CHART 34.



I.—Charts showing that Auto-inoculation Effects are Elicited by Operative Interference with Foci of Bacterial Infection.

Chart 34 refers to a child, aged seven rs, who was admitted to St. Mary's spital with caries of the fibula. Up the time of admission the child had n running about in the country. Her tuberculo-opsonic index king out at 1·4, we came to the conclusion that the child had erculous disease, that she had auto-inoculated herself, that immunisa-

tion was in progress, and that it would be advisable to postpone vaccin therapy until such time as the child's tuberculo-opsonic index shoul have returned to the normal. Coming back to the case a few days aft the patient's blood had been found to be normal, we found that a scrapir operation had two days previously been undertaken, and that the patient index had risen to 1·7, to fall off again on the next day to 1·15. Aft watching the progress of events for another fortnight and finding that th patient's index was no longer fluctuating and that the case was nc making rapid progress, we proceeded on October 18 to inoculate wit tuberculin. On returning to test the result of this inoculation we foun that a second scraping operation had been undertaken and that th rise of the opsonic index had been checked. Continuing our measur ments of the opsonic index we regis tered, as will be seen in the curve, typical negative and positive phas Finally, when the patient's index ha returned to the normal, we resume our tuberculin inoculations.

CHART 35.

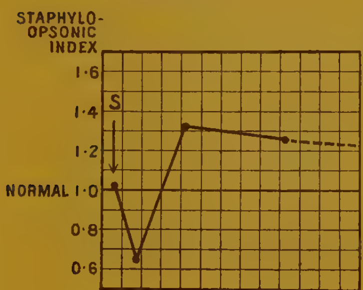


Chart 35 refers to a boy, age twelve years, who was admitted to S Mary's Hospital with osteomyeliti The curve shows the typical nega tive and positive phase which supe vened upon the opening up and scra ing of the focus of infection.

Chart 36 refers to a child, aged seven years, with tuberculous diseas of the hip, and *Chart* 37 to a child with tuberculous glands on both side

CHART 36.

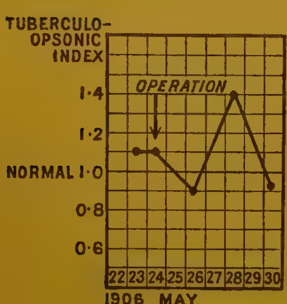
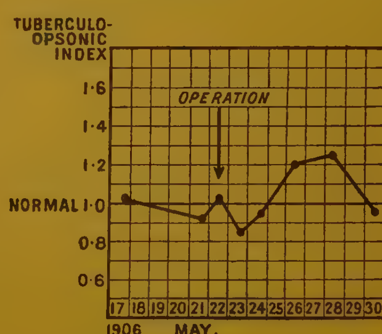


CHART 37.



of the neck. The curves show that in each case a typical fluctuation c the tuberculo-opsonic index supervened upon operation.

.—Charts showing that Auto-inoculation Effects are obtained by Active and Passive Hyperaemia affecting Foci of Bacterial Infection.

Chart 38 relates to a patient, a boy, aged sixteen years, who pre-nted himself for treatment with an extensive ulcer on the front of the leg elow the knee standing in connexion with carious bone in the head of ie tibia. The patient was taken into hospital and his blood was drawn f for examination, once on November 20, twice on the 21st, and again on ie 22nd. His tuberculo-opsonic indices worked out for these occasions as ·78, 0·7, 0·7, and 0·8 respectively. At 3 p.m. on the 22nd, immediately fter the sample of blood which gave the reading last mentioned had been ithdrawn, a hot poultice was applied to the ulcer. At 4 p.m., when the oultice was removed, the patient's tuberculo-opsonic index stood at ·87; it stood at 6 p.m. at 1·1; at 8 p.m. at 1·06; and at 10 p.m., at 0·9. n the 23rd, at 2 a.m. it stood at 0·94; at 6 a.m. at 1·12; at 12 noon at ·43; and at 6 p.m. at 1·35. Finally, on the 24th, the patient's index ad reverted to its normal of 0·75.

Manifestly we have here a quite typical auto-inoculation effect pro-uced by bacterial products washed out of the focus of infection by the mpler lymph flow which is obtained by active dilatation of the blood essels. (Cf. Table, Serial Number 17.)

Chart 38.

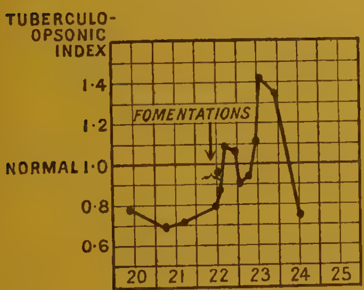
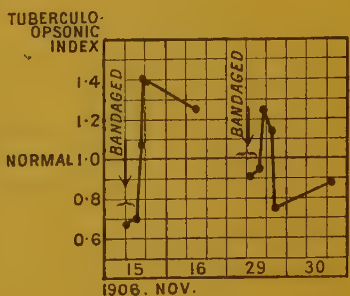


Chart 39.



The series of observations recorded in Charts 39–43 were made by us onjointly with Dr. F. S. Patch, of Montreal. They show that auto-noculation effects are induced by the passive hyperaemia which is re-ommended by Bier. Here, as in the case of active hyperaemia, the ause of the blood changes must assuredly be sought in the washing out f bacterial products from the focus of infection by the agency of the ccelerated lymph stream.

Chart 39 refers to a young woman who was suffering from extensive upus of the face and of both elbows. In the first of the two observations vhich are in question in this chart the bandage was applied to the right pper arm for a period of two hours, and readings of the tuberculo-psonic index were taken, immediately before applying the bandage,

immediately after the removal of the bandage, again two and four hou afterwards, and finally twenty-four hours afterwards. On the secor occasion, some fourteen days later, the bandage was applied round t arm for an hour, readings of the tuberculo-opsonic index being tak immediately before bandaging, half an hour after the bandage had be applied, immediately after the removal of the bandage, one hour afte six hours after, and again twenty-four hours after. A striking clinic amelioration was here obtained from the application of the bandage.

Chart 40 refers to a boy, aged ten years, who was suffering from lup of the right hand associated with superficial tuberculides in both fee The bandage was applied round the forearm. On the two occasions c

CHART 40.

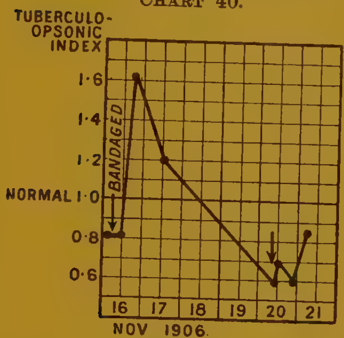
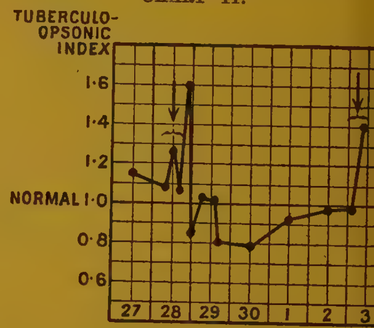


CHART 41.



which it was employed it was applied in each case for one hour, reading of the tuberculo-opsonic power being taken immediately before bandagin immediately after bandaging, and again eight and twenty-four hour after bandaging.

Of incidental interest in connexion with this chart is, first, the fact tha we have here an example of the secondary ebb to which attention wa directed at the outset of this paper, and further, the fact that a dispro portionately small result, as compared with that obtained on the firs occasion, was here registered in association with the second applicatio of the bandage.

CHART 42.

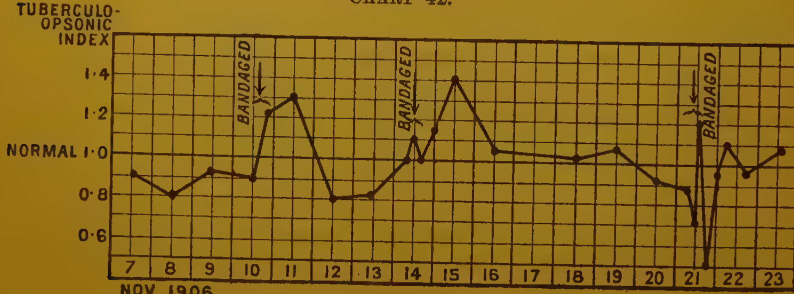
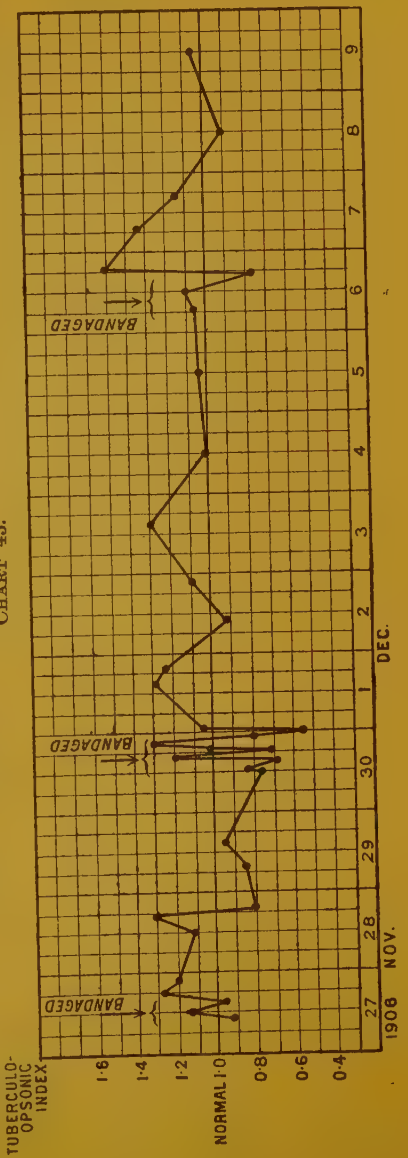


Chart 41 refers to a girl, aged fifteen years, who was the subject of tuber-lous synovitis of the right knee. In the first of the two observations iich are in question in this chart the bandage was applied for one and a lf hours, readings of the tuberculo-sonic index being taken twenty-ur hours before, immediately fore, in the mid-period of the ndaging, immediately after the ndaging, again one hour, six hours, ghteen hours, and again thirty urs after the bandaging, and then ily for a succession of days. In e case of the second observation adings were taken only at the begin-ng and at the end of the bandaging.

Chart 42 applies to a child, aged vo and a half years, who was affect-l with tuberculous synovitis of the ght knee. Three separate observa-ons were here made in the course of fortnight, the patient being kept in ed during the whole period] with a oroplastic case applied to the limb. he bandage was applied, on the rst occasion, lightly for three hours, nd on the second occasion equally ghtly for three and a half hours.)n the third occasion it was applied nuch more tightly, but only for one nd three-quarter hours. On this ccasion one reading of the tuber-ulo-opsonic index was taken imme-liately before the bandaging, three eadings during the period of band-ging, and again another six hours after the removal of the bandage.

Chart 43 refers to a boy, aged nine years, who was affected with exten-sive glandular and cutaneous tuber-culosis and who had undergone a very large number of operations with a view to the relief of these conditions. Large ulcers occupied the left fore-arm, the dorsum of the left hand, and the right supraclavicular region. Three separate bandaging observations are in question in the chart, the bandage being applied on each occasion

CHART 43.



round the upper arm on the left side. It was applied on the first occasio for one hour, and on the second and third occasions in each case for thre hours.

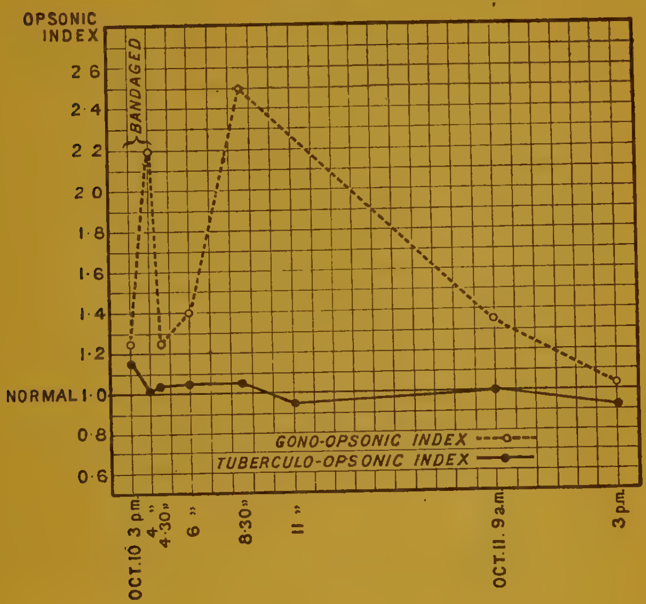
Of interest in connexion with this case are the following points:—(a Very striking amelioration of the patient's condition was here obtaine the open ulcers all healing over in the interval between the first an third bandaging. (*b*) The amelioration was not confined to the bandage left limb; it was almost equally conspicuous in the large ulcer whic occupied the supraclavicular region on the right side of the body. (c On the first and second occasions on which the bandage was applie lymph poured out in a copious stream from open ulcers on the dorsum o the hand and forearm—showing clearly that we have to deal in the cas of the bandaged limb with an activated lymph flow. (*d*) The lymph whicl poured out from the ulcer gave on each occasion lower tuberculo-opsoni readings than the circulating blood. In the case of the first bandagin the two samples of lymph which were tested gave readings of 0·8 and 0·6 respectively; the six successive samples of lymph which were tested or the second occasion gave a series of readings ranging downwards by gradual steps from 0·7 to 0·2. It may, we think, be inferred from this fact that the opsonic index of the fluid in the interior of the tuberculous focus stood at 0·2, or even lower, at a time when the opsonic index of the circulating blood stood at 0·8. (*e*) Finally, it is of interest to mention that on a subsequent occasion, when after a relapse the bandage was again applied, a lymph was obtained which conveyed tuberculous infection to a guinea-pig.

PART II.

Chart 44 refers to a patient who presented herself for treatment with a swollen wrist-joint which was suspected to be tuberculous. It appeared that the patient had in the past suffered from arthritis in the left hip and the right shoulder, and that these had been attributed to gout. The patient's tuberculo-opsonic index was measured on September 25 last and on October 4 and 8. It worked out on the first occasion as 0·94, on the second as 0·95 and on the third as 0·97. As these readings furnished no indications of any tuberculous auto-inoculation, the patient was further questioned and a history of vaginal discharge was elicited. This discharge had, however, completely ceased, and the vaginal secretion proved to be free from gonococci. The patient's gono-opsonic index was measured on October 4, 7, and 8. The readings obtained on these occasions were 1·1, 1·35, and 1·03. As these readings furnish no indication, or at best only a very doubtful indication, of a gonococcal auto-

oculation, a Bier's bandage was now applied to the patient's forearm
one hour. The gono- and tuberculo-opsonic readings which were
tained in association with this procedure are set out in the chart. It

CHART 44.



ll be seen that they show a characteristic fluctuation in the gono-
sonic index, while the tuberculo-opsonic index remains quite unaffected.
view of this result the patient is now being treated with gonococcus
ccine.

Table illustrating some of the Diagnostic and Therapeutic Problems that can be resolved by Recourse to the Auto-Inoculation Test and by Consideration of its Event.

Serial Number.	Clinical Features of Case.	Diagnostic or Therapeutic Problem.	Procedure resorted to with a view to the Induction of an Auto-inoculation.	Particulars of Opsonic Readings.	Conclusion arrived at, and Remarks.
				Tuberculo-opsonic Indices.	
1	Tuberculous phthisis. Tubercle bacilli in sputum	May patient take gentle exercise ?	30 minutes' sharp walk	Before walk . . . . 1·09 Immediately after . . 1·02 6 hours after . . . 1·06 24 ,, ,, . . . 0·98	Yes ; he may take gentle exercise.
2	Same patient as No. 1	May he take hard exercise ?	70 minutes' hard walk	Before walk . . . . 0·92 Immediately after . . 0·96 6 hours after . . . 1·14 24 ,, ,, . . . 1·03	Since 70 minutes' hard walk appears to induce an auto-inoculation it will probably be well for him to avoid very severe exercise.
3	Tuberculous disease of knee-joint . .	Should vaccine-therapy with tuberculin be continued ?	Walking exercise	Before walking . . . 1·10 Immediately after. . 1·19 5 hours after . . . 1·03 9½ ,, ,, . . . 1·41 24 ,, ,, . . . 1·32	Tuberculin inoculations ought to be continued. (*Vide serial number 37 infra.*)
4	Patient was suspected of phthisis and was rejected for insurance, but was declared sound by two independent physicians.	Was he rightly rejected ? (First test)	Vigorous walk	Before walk . . . . 0·85 Immediately after . . 0·62 6 hours after . . . 0·73 24 ,, ,, . . . 0·80	Patient is tuberculous and was rightly rejected.
5	Same patient as No. 4	(Second test)	,, ,,	Before walk . . . . 0·79 Immediately after . . 0·56 6 hours after . . . 0·71 24 ,, ,, . . . 0·63	
6	Tuberculous disease of the knee . .	May the knee be massaged to obtain more freedom of movement ?	Massage of affected joint	Before massage . . . 0·91 1 hour after . . . 1·44 6 hours after . . . 0·76 7 days ,, . . . 1·22	Knee must be kept at rest and inoculation with tuberculin would be appropriate.
7	Chronic arthritis of ankle-joint. In Australia lesion was diagnosed by one physician as rheumatic arthritis, by another, who found that his tuberculo-opsonic index was 0·74 as probably tuberculous.	Is the arthritis rheumatic or tuberculous ?	Massage of affected joint	Before massage . . . 0·76 Immediately after . . 0·83 6 hours after . . . 0·97 24 ,, ,, . . . 1·00 48 0·74	Patient is suffering from tuberculous arthritis. Inoculations with tuberculin are indicated.—These are being carried out with marked advantage.

				6 hours after	1·02	tuberculin are indicated.—These are being carried out with marked benefit to the patient.
				24 „ „	0·93	
9	Illness commencing acutely with abdominal symptoms and high fever. Abdominal pain and tenderness continuing, tuberculous peritonitis is suspected	Is the disease tuberculous ?	Gentle massage of abdomen for 10 minutes	Before massage	1·31	The disease is tuberculous.—The patient recovered completely under tuberculin treatment.
				Immediately after	1·12	
				6 hours after	0·88	
				36 „ „	1·76	
10	Suspected tuberculous disease of hip	Is the disease tuberculous ?	Movement of affected joint	Before movement	0·90	The diagnosis still remains doubtful.
				½ hour after	1·16	
				2½ hours after	1·02	
				7½ „ „	0·94	
11	Since sprain eight months ago swelling and pain in left wrist, which has been getting progressively worse	Is the disease tuberculous ?	Bier's bandage applied to fore-arm of affected side for half an hour	Before bandaging	0·74	The disease is probably tuberculous.
				Immediately after	0·92	
				¾ hour after	0·89	
				18 hours after	0·77	
				36 „ „	0·81	
12	Arthritis of ankle associated with much swelling, some tenderness, and pain. The onset was acute. A skiagram showed some bony lesion in tarsus. Amputation had been advised in another hospital, but patient refused operation	Is the disease tuberculous ?	Bier's bandage applied above ankle for one hour	Before bandaging	0·92	The disease is tuberculous.—Inoculations of tuberculin have already produced a most marked improvement in the condition.
				Immediately after	0·81	
				4 hours after	1·00	
				24 „ „	0·74	
13	Painless swellings of the left wrist, elbow, knee, and ankle of three months' duration. Knee had been tapped and fluid had furnished a culture of staphylococcus albus. Sinus in ankle yields a culture of staphylococcus	Is the arthritis tuberculous or staphylococcic ?	Wrist massaged for ten minutes.	Before massage	0·98	The disease appears to be neither tuberculous nor staphylococcic.
				½ hour after	1·01	
				4½ hours after	1·15	
				22 „ „	1·08	
				30 „ „	1·15	
				Staphylo-opsonic Indices.		
				Before	0·70	
				½ hour after	0·88	
				4½ hours after	0·78	
				22 „ „	—	
				30 „ „	0·78	
				Tuberculo-opsonic Indices.		
14	Tuberculous arthritis of hip with secondary infection and sinuses which had been treated by inoculation.	Is the tuberculous disease eradicated ?	Vigorous movement of hip-joint	Before massage	1·06	Tuberculous disease is still present.
				¾ hour after	0·96	
				2 hours after	0·86	
				5½ „ „	0·85	
				12 „ „	1·06	
				24 „ „	1·25	
				3 days „	0·69	

Table illustrating some of the Diagnostic and Therapeutic Problems that can be Resolved by Recourse to the Auto-Inoculation Test and by Consideration of its Event—(continued).

Serial Number.	Clinical Features of Case.	Diagnostic or Therapeutic Problem.	Procedure resorted to with a view to the Induction of an Auto-inoculation.	Particulars of Opsonic Readings.	Conclusion arrived at, and Remarks.
15	Dactylitis of two fingers following fall from a horse	Is disease tuberculous ?	Massage of affected fingers	Before . . . . . 0·95 ½ hour after . . . 0·91 5½ hours ,, . . . 0·73	The disease probably is tuberculous.—Practically complete recovery has taken place under tuberculin treatment.
16	History of swelling of joints after strain. Twice treated in hospital abroad for gonococcal arthritis	Is disease due to gonococcus ?	Massage of affected joint	*Gono-opsonic Indices.* Before massage . . . 0·97 6 hours after . . . 1·00 24 ,, ,, . . . 1·04	The arthritis is not of gonococcal origin.
17	Tuberculous arthritis of knee and tuberculous kidney	Does a hot fomentation applied to a tuberculous lesion produce an auto-inoculation ?	Application of hot fomentations to tuberculous knee	*Tuberculo-opsonic Indices.* Before fomentation. . 1·00 1 hour after . . . 1·32 2 hours ,, . . . 1·14 8 ,, ,, . . . 1·14	Yes. (*Cf. also Chart* 34.)
18	Tuberculous arthritis of wrist . .	May tuberculin inoculations be abandoned ?	Massage of joint and passive movement	Before massage . . . 0·78 15 hours after . . . 1·24 24 ,, ,, . . . 1·06 48 ,, ,, . . . 1·17	No. Tuberculin inoculations must continue.
19	Ganglion on dorsum of wrist occurring in a patient with other tuberculous lesions	Is this ganglion tuberculous ?	Massage of ganglion	Before massage . . . 0·65 After ,, . . . 0·65 1½ hours after . . . 0·69	Apparently not.
20	Tuberculous glands removed from neck five years ago. Diffuse swelling of wrist following accident five months ago	Is the swelling of wrist tuberculous in character ?	Massage and movement of wrist for 15 minutes	Before massage . . . 0·98 After ,, . . . 0·76 24 hours after . . . 0·74	Yes.
21	Uncomplicated tuberculous hip treated by complete rest and inoculations of tuberculin for five months	May inoculations cease?	15 minutes massage	24 hours before . . 0·85 Immediately before . 0·91 ,, after . . 0·80 3 hours after . . . 0·90 15 ,, ,, . . . 0·94 24 ,, ,, . . . 1·10 36 ,, ,, . . . 0·81	Yes.

22	Arthritis of long standing in a patient who has had gonorrhoea	Is arthritis gonorrhoeal ?	Massage of affected joint	*Gono-opsonic Indices.* Before massage . . . . 1·10 15 hours after . . . 1·26 24 " " . . . 1·37 3 days " . . . 1·02 4 " " . . . 1·00	The arthritis is probably gonorrhoeal.
23	Tuberculous disease of ankle . (See p. 415 *supra*, Chart 28.)	Is tuberculous focus extinct ?	Walking on affected ankle without splints	*Tuberculo-opsonic Indices.* Before walking . . . 0·83 Immediately after . . 0·80 12 hours after . . . 0·67 24 " " . . . . 0·79	Tuberculous disease may still be present.—Tuberculin inoculations were continued.
24	Same patient as 23, 26 weeks later .	Is tuberculous focus extinct ?	Walking and bicycling	Before . . . . . . 0·85 After . . . . 0·73 12 hours after . . . 0·73 24 " " . . . . 0·84	Probably focus is extinct.
25	Tumour in hypogastrium in connexion with the uterine body diagnosed as probably tuberculous;	Is tumour tuberculous ?	Massage of tumour for 20 minutes	7 days before massage. 0·83 3 " " " . 0·87 Immediately before . 1·23 " after . . 1·25 5 hours after . . . 1·30 24 " " . . . . 0·96	Tumour is tuberculous.—Tumour has markedly shrunk under the influence of tuberculin inoculations.
26	History of pain in hip and lameness. Examination by surgeon gave a negative result. Patient sent on for corroboration or otherwise of this diagnosis	Is disease tuberculous, or may boy return to school ?	Vigorous massage and movement of hip ;	Before massage . . . 1·00 Immediately after . . 1·09 6 hours after . . . 1·05	Disease is not tuberculous and boy may return to school.
27	Large mass of glands in groin, followed by development of extensive nodular masses in abdomen	Are the glands tuberculous or malignant ?	Glands vigorously massaged	*Tuberculo-opsonic Indices.* Before massage . . . 0·96 Immediately after . . 1·45 6 hours after . . . 0·95 24 " " . . . . 1·70 *Neo-opsonic Indices.* Before massage . . . 1·02 Immediately after . . 1·02 6 hours after . . . 0·95 24 " " . . . . 1·67	Patient is suffering from tuberculous glands and malignant disease. Patient died some weeks later and at the necropsy it was found that the pelvic peritoneum was infiltrated with lymph-adenomatous masses. The retro-peritoneal and mesenteric glands were all enlarged and in some definitely caseated tuberculous deposits were found. (*Vide Chart* 27.)

Table illustrating some of the Diagnostic and Therapeutic Problems that can be Resolved by Recourse to the Auto-inoculation Test and by Consideration of its Event—(continued).

Serial Number.	Clinical Features of Case.	Diagnostic or Therapeutic Problem.	Procedure resorted to with a view to the Induction of an Auto-inoculation.	Particulars of Opsonic Readings.	Conclusion arrived at, and Remarks.
28	Suspected tuberculous laryngitis. Patient sent up from out-patient department for diagnosis	Is laryngitis tuberculous ?	Talked vigorously for one hour	*Tuberculo-opsonic Indices.* 24 hours before . . 0·94 18 " " . . . 0·98 Immediately before . 0·87 " after . . 1·00 ¼ hour after . . . 1·12 3¼ hours " . . . 1·24 6¼ " " . . . 0·83 8¼ " " . . . 1·00 12 " " . . . 0·88 20 " " . . . 1·00 30 " " . . . 1·00 2 days " . . . 0·75 3 " " . . . 0·75	Laryngitis is probably tuberculous.
29	Osteomyelitis of femur with sinus. Sinus has persisted for seven years	Is there here persistent staphylococcic infection ?	Bier's bandage applied for one hour	*Staphylo-opsonic Indices.* Before bandaging . . 0·53 During " . . 0·81 Immediately after . . 0·72 ¼ hour after . . . 0·84 1¼ hours " . . . 0·93 2¼ " " . . . 1·10 36 " " . . . 1·81 72 " " . . . 0·62	There is here persistent staphylococcic infection.—The sinus completely healed under the influence of staphylococcic inoculation.
30	Affection of eye diagnosed as probably tuberculous; suspected tuberculous disease of lung	Is pulmonary tuberculosis present ?	Vigorous tennis playing	*Tuberculo-opsonic Indices.* 24 hours before playing 0·65 Immediately before . 0·75 " after . . 0·75 5 hours after . . . 0·99 24 " " . . . 1·00	Pulmonary tubercle is probably present.

			knee	8 hours after . . . 24 " " . . . 30 " " . . .	0·49 0·72 1·05	that the fluid that was withdrawn was injected into a guinea-pig without rendering it tubercular supported the idea that we had here a non-tubercular arthritis. Subsequent inquiry revealed that the anaesthetist had at the operation detected signs of tubercular infection at one apex.
32	Polyarthritis (both knees, both ankles and fingers)	Is the arthritis tubercular ?	Bier's bandages applied above both knees	24 hrs. before bandaging Immediately before . 1 hour after . . . 8 hours " . . . 24 " " . . . 48 " " . . . 72 " " . . .	0·82 1·08 1·08 0·87 1·26 1·03 0·89	The arthritis is tubercular.
33	Pulmonary tuberculosis	Does exercise here cause an auto-inoculation ?	Ordinary exercise	Morning . . . . . Evening	1·06 1·22	
34	Tubercular disease of the knee . .	Has the tubercular infection been extinguished by vaccine-therapy, and may the patient lay aside his splints ?	Walking	Before walk . . . Immediately after . . ½ hour after . . . 2½ hours " . . . 6½ " " . . . 16 " " . . . 20 " " . . .	1·28* 1·18 1·20 1·13 0·95 1·17 1·35	The tubercular infection has here been almost extinguished. The patient who before this investigation was undertaken had been pronounced by the surgeon in charge to be free from all objective signs of disease, was authorized to discard his splints, but was kept under observation for another four months. After the lapse of another two months, during which time he remained perfectly well, patient contracted the infection of influenza. In association with this the symptoms of tubercular disease manifested themselves again in the knee, and a tubercular nodule developed in the ankle.
35	Tubercular disease of the hip . . .	Has the tubercular infection been extinguished by vaccine-therapy, and may the patient discard his crutches ?	Walking	Before walk . . . Immediately after . . 1 hour after . . . 3 hours " . . . 5 " " . . . 10 " " . . . 20 " " . . .	1·02 0·95 0·62 0·94 1·17 1·12 1·2	The tubercular infection still persists; patient must not discard his crutches, and inoculations must be continued. The patient who before this investigation was undertaken had by the surgeon in charge been pronounced free from all objective signs of disease suffered thereafter from a return of all his symptoms.

* This high index is no doubt due to a tuberculin inoculation which the patient had received ten days previously.

Table illustrating some of the Diagnostic and Therapeutic Problems that can be resolved by Recourse to the Auto-Inoculation Tests and by Consideration of its Event—(continued).

Serial Number.	Clinical Features of Case.	Diagnostic or Therapeutic Problem.	Procedure resorted to with a view to the Induction of an Auto-inoculation.	Particulars of Opsonic Readings.	Conclusion arrived at, and Remarks.
[illegible]	Arthritis with effusion of left knee diagnosed as tubercular	Is the infection tubercular?	Bier's bandage applied above the knee	Before bandaging . . 1·06 ½ hour after . . . 0·89 5 hours " . . . 0·9 20 " " . . . 1·0	The arthritis is not tubercular.
		Is the infection gonococcal?	Same application as was in question above	*Gono-opsonic Indices.* Before bandaging . . 1·4 ½ hour after . . . 1·24 5 hours " . . . 1·36 20 " " . . . 1·84	The arthritis is gonococcal.—Subsequent inquiry revealed that the patient had suffered from a vaginal discharge six weeks before the arthritis had manifested itself, and that she had afterwards suffered from a contracture of the hand.
37	Tuberculous disease of knee joint. Same case as No. 3. After three months' further tuberculin treatment	Has the tubercular infection been extinguished?	Walking exercise	*Tuberculo-opsonic Indices.* Before walk . . . 0·93 After " . . . . . 1·10 8 hours after . . . 0·96 24 " " . . . 1·00 48 " " . . . 0·98	The tuberculous focus is probably extinct. The patient has been doing full day's work for the last six months without any recurrence.
38	Disease of tarsus, probably tuberculous	Is condition tuberculous?	Exposure to X Rays	3 hours before . . 0·73 1 hour " . . 0·74 ½ hour after . . . 0·84 5½ hours " . . . 0·60 20 " " . . . 0·62	Condition is tuberculous as shown by the persistent low index, but the exposure to X rays was insufficient to induce an auto-inoculation.
39	Arthritis of knee.	Is condition tuberculous?	Bier's bandage applied ¾ hour	Before . . . . . . 1·08 ½ hour after . . . 1·35 3 hours " . . . 1·16 6 " " . . . 1·12 18 " " . . . 0·93	Condition is tuberculous.
40	Arthritis of knee.	Is condition tuberculous?	Bier's bandage applied ¾ hour	Before . . . . . . 1·02 Immediately after . . 1·11 6 hours after . . . 1·27 18 " " . . . 1·16	Condition is tuberculous.

41	Suspected tuberculous peritonitis	Is condition tuberculous?	Massage of abdomen	Before	[illegible]	Condition is tuberculous.
				½ hour after	1·25	
				3 hours "	1·09	
				8 " "	1·82	
				24 " "	0·83	
42	Pulmonary tuberculosis, bacilli in sputum	Does hard walking induce an auto-inoculation?	Eight mile walk	Before walk	0·61	Walk not sufficient to induce auto-inoculation.
				After "	0·63	
				2 hours after	0·62	
43	Suspected tuberculous infection	Is a tuberculous infection present?	Vigorous dancing	Before dance	1·11	A tuberculous infection is present.
				Immediately after	1·25	
44	Same case as Nos. 43 and 45	Is a tuberculous infection present?	Second dance (the day after the previous one)	Before dance	1·58*	A tuberculous infection is present, and it would be unwise for patient to dance two days in succession.
				After "	1·14	
				10 hours after	0·77	
45	Same case as Nos. 43 and 44	Is a tuberculous infection present?	Vigorous golfing exercise	Before	1·23	A tuberculous infection is present.
				After	1·18	
				6 hours after	1·08	
				24 " "	1·36	
46	Suspected tuberculous meningitis	Is a tuberculous infection present?	Lumbar puncture. 20 c.c. fluid withdrawn	Before	0·84	The condition is one of tuberculous meningitis. This was confirmed at the autopsy.
				5 hours after	1·23	
				24 " "	1·16	
				3 days "	0·88	
47	Acute pleurisy	Is this pleurisy tuberculous?	Vigorous rubbing of chest with liniment	24 hours before	0·92	Pleurisy is tuberculous.
				12 " "	1·08	
				5 " after	1·50	
				17 " "	1·23	
				24 " "	1·12	
48	Pulmonary Phthisis	Does a long walk cause an auto-inoculation?	Two hours' walk (8 miles)	Before	0·85	Walking is sufficient to induce an auto-inoculation.
				After	0·72	
				3 hours after	1·15	
				6 " "	1.21	
49	Same case as No. 48 after tuberculin treatment	Same problem (six months later)	Two hours' walk (same miles)	Before	0·92	Walking does not now induce an appreciable auto-inoculation.
				After	0·84	
				4 hours after	1·03	
				7 " "	0·98	
50	Ulceration of both legs	Is condition tuberculous?	Bier's bandage, both legs	Before	0·74	Condition is tuberculous.
				½ hour after	1·03	
				4½ hours after	1·06	
				18 " "	0·64	

* This result is no doubt due to the auto-inoculation due to the dancing of the previous day.

On some Points in connexion with Vaccine-Therapy and Therapeutic Immunisation generally.[1]

Being the substance of a Lecture delivered before the Harveian Societ[y] of London, March, 1908.

By A. E. Wright.

Introductory

Part I.—Question as to whether it would not be possible to achieve and maintain an in-creased Output of protective substances apart from Periodic Measurements of th[e] content of the Blood in Protective Substances—Suggestion that by Clinical Observa-tions and Blood Testings such a Knowledge of the Effects of bacterial vaccine[s] has been arrived at as would make it possible to guarantee a good Result fron[m] the Inoculation of a definite Quantum of a Standardised vaccine—Suggestion tha[t] the clinical symptoms of the patient will furnish to the immunisator a guid[e] by which he may regulate his immunisation procedures—Résumé of the Con-clusions which have been arrived at—Does the technique for the Determinatio[n] of the opsonic index furnish to the immunisator accurate and useful infor-mation with respect to the way in which the patient is progressing?

Part II.—How may such antibacterial agencies as the patient may already be possesse[d] of or may acquire by immunisation, be directed to the destruction of microbes i[n] the focus of infection?—General question of the distribution of antibacteria[l] agents in the Normal Organism—Nature of the inflammatory reaction whic[h] supervenes upon a bacterial invasion of the tissues, and changes in th[e] Distribution of the Antibacterial Agents which are effected by that Reaction—Conditions which present themselves in the Case where Microbes have survive[d] the Inflammatory Reaction, which has supervened upon their Invasion, an[d] have established themselves in a Nidus in the Tissues—Consideration of th[e] Therapeutic Measures by which we may bring the Antibacterial Agencies of th[e] Circulating Blood into Effective Operation upon the Microbes in a Nidus o[f] Infection—Organization of the Medical Profession for carrying out the Work o[f] Therapeutic Immunisation—Conclusion.

Already in my first paper on the treatment of staphylococcus infection[s] by the therapeutic inoculation of staphylococcus vaccines,[2] I suggeste[d] that we had in vaccine-therapy a general therapeutic method whic[h] would be applicable to the treatment of all kinds of localized bacteria[l] infections.

[1] Reprinted from the *Practitioner.* Special Number on "The Opsonic Method and Vaccine-therapy," May, 1908.

[2] *Lancet,* March 29, 1902, *vide* pp. 224–226.

n my next publication on therapeutic inoculation, I made bold to dict that the physician of the future would be an immunisator.[1]

lready these anticipations are justifying themselves. I do not w that there is any one who has made trial of vaccine-therapy connexion with localized bacterial diseases who is not satisfied with efficacy as a therapeutic measure, and the day when the physician be an immunisator is, I think, perceptibly nearer.

Far from the possibilities of vaccine-therapy in the field of the treat- nt of localized bacterial infections being already exhausted, those sibilities have as yet been only very incompletely explored. Only a and tentative experiments have, up to the present, been undertaken connexion with bacterial infections of the mucous membranes. The plication of vaccine-therapy in connexion with endometritis and bron- tis is almost unstudied, and the method has not yet been applied to ooping cough or to mumps. The question as to whether the Klebs- effler bacilli, in the case where these survive on the throat after an ack of diphtheria, can be eradicated by vaccine-therapy, and the cisely similar question which presents itself in connexion with the sistence of typhoid bacilli and other pathogenetic bacteria after con- escence, are in like manner still untouched. Again, while it may taken as certain that in malignant disease advantage may be obtained m vaccine-therapy directed to the destruction of the microbes which ade the tumour, final judgment on the question whether this has retarding effect upon the disease cannot yet be pronounced. Lastly, cannot be doubted that there are numberless disorders which, though y are not now brought into connexion with bacterial infection, are reality directly caused by such. Many of these might prove amenable vaccine-therapy. It has been suggested to me that vaccine-therapy ght perhaps with advantage be employed in connexion with hay fever. might also be successfully applied in dentistry in connexion with the ief of toothache in the same way as it has already been applied to the atment of pyorrhoea alveolaris. Again, jaundice and cholecystitis ght be treated by vaccine-therapy, and if I may draw any deduction m an almost desperate case, which I have recently seen get well under ccine-therapy, there would be a prospect of success in certain cases pancreatitis. And I cannot refrain from throwing it out as a ggestion that there may quite well lie at the root of pancreatic betes a bacterial infection (possibly sometimes a colic or a tubercle ection) which might perhaps be amenable to vaccine-therapy.

Leaving all these problems for the future to resolve, I may, before pass to the subject-matter proper of this paper, put you in mind of tain recent developments in connexion with vaccine-therapy and to-immunisation.

(*a*) It has been established by my fellow-workers and myself in the

[1] *British Medical Journal*, May 9, 1903, *vide* p. 227.

Inoculation Department of St. Mary's Hospital, that vaccine-therapy i
applicable not only to localized infections but also to septicaemic diseases

(*b*) We have shown that spontaneous auto-inoculations occur no only in connexion with septicaemic diseases, but also in connexion wit localized diseases where the focus of infection has attained to a certai development.

(*c*) We have shown that auto-inoculation can, in the case c localized disease, be artificially induced by massage, by active an passive movements, and by active and passive hyperaemia affectin the focus of infection.

(*d*) We have shown that such artificially-induced auto-inoculation can be turned to useful account in diagnosis, evidence of an auto-inocula tion with the products of a particular microbe being equivalent to proo of the presence of that microbe in the particular region of the bod which has been explored.

(*e*) We have shown that the success of certain empirical method of treatment, and, in particular, the success of Bier's method of passiv congestion, is probably largely dependent upon the fact that, whe skilfully and felicitously applied, it induces adequate and not excessiv auto-inoculations.

I do not propose here to go back over any of this ground. It ha been traversed by my fellow-workers and myself in a conjoint pape contributed to the *Lancet* of November 2nd last.[1]

Let me, instead, deal with two other issues which present themselve for consideration in connexion with therapeutic immunisation.

The first of these has reference to the question as to whether it woul not be possible to achieve and maintain the desired improvement in th patient's blood apart from periodic blood-examinations. Associated wit this is the question as to whether the technique, which we use for the determina tion of the opsonic index, gives accurate and useful results.

The second issue has to deal with the question as to what is the best metho of directing such protective agencies, as a patient may be possessed of, o acquire by immunisation, to the destruction of the microbes in the focu of infection.

PART I

Question as to whether it would not be possible to achieve and maintai an increased output of protective substances apart from periodi measurements of the content of the blood in protective substances.

The importance of this issue will appear when we reflect upon wha is bound up with it. There is bound up with it the question whethe

[1] *Vide* pp. 372–429.

s imperative upon every physician, before embarking upon therapeutic munisation, to serve an apprenticeship in bacteriology, to master e technique of blood testing, and thereafter to undertake, in connexion th his cases, the labour of making periodic blood examinations.

In discussing this question, I think we may, at this hour of the day, nfidently assume that no one would now seriously advocate that ections of bacterial vaccines should be undertaken entirely in the rk, as was done in connexion with the earlier inoculations of Koch's berculin. Every one will agree that since we have, in vaccines, an ency that is, according as it is wisely or unwisely used, powerful good or for ill, we must, where we employ vaccines, either have a arantee for the correctness of the dosage, or a system of control which ll tell us when our dose is too large, when it is too small, and when it ght to be repeated. I may, therefore, take it that the suggestion at the inoculation of bacterial vaccines should be undertaken apart om anything in the nature of controlling blood examinations, is to understood not as a contention that the dosage need not be a matter concern, but rather as a suggestion that the accumulated experience the past, and the observation of the clinical symptoms, will serve adequate guides in any immunisation procedures.

This suggestion—though it may perhaps have suffered something om the advocacy of those who are above everything concerned to oid anything in the nature of undue personal effort, and anything the nature of a change in the established order in our profession—is ne the less deserving of our most earnest consideration, for there n be no question but that the shortest and least laborious way to a sult is, where it can be safely followed, always the best way.

It will facilitate the discussion of the question as to how far periodical ood examinations may be dispensed with in favour of other methods of gulating the dosage, if you will allow me to introduce a little more pre-ion into the suggestion that past experience and observation of the nical symptoms may serve us as a sufficient guide into our immunisation ocedures. We may read this (*a*) as a suggestion that we have already, clinical observation and blood testings, attained to such knowledge of e effects of bacterial vaccines as enables us to guarantee that a good sult will follow upon the inoculation of a particular dose of a standard-d vaccine. Or, again, we read it (*b*) as a suggestion that, even if such solute guarantee as might be desirable should not yet be available, e clinical symptoms of the patient will inevitably furnish such rther data as might be required for the regulation of his dosage.

ggestion that by clinical observations and blood testings such a knowledge of the effects of bacterial vaccines has been arrived at as would make it possible to guarantee a good result from the inoculation of a definite quantum of a standardised vaccine.

Though inexperience might suppose that the blood testings, which

have been carried out, and the clinical experience which has been gained would enable us to foretell with absolute accuracy the effect of a given dose of this or that vaccine upon the human organism, we have not only not arrived at this point, but there is very little likelihood of our ever arriving at it. Where we are handling, on the one hand, vaccines and on the other hand, the human organism, we are dealing with factors neither constant nor invariable.

Let us consider, first, the vaccine. Here, even where we particularise a particular dose of a vaccine which has been standardised by counting the contained microbes or by weighing the contained microbial substance, we can never leave out of consideration the possibility that a difference in the strain of microbes employed, or some minute overlooked difference in the mode of preparation, or perhaps some change occurring spontaneously during keeping, might affect the potency of the vaccine. It will follow that where we prescribe, let us say, a dose of gonococcus or streptococcus vaccine, such as would correspond to 1,000,000 gonococci, or, as the case may be, 1,000,000 streptococci, we cannot always be sure that we shall be applying precisely the same *ictus immunisatorius*. Here, then, is one of the difficulties which stand in the way of our accurately forecasting the effect of the inoculation of a bacterial vaccine. It is a difficulty which can be circumvented only by directly controlling the effect of the vaccine upon the patient. More formidable is the difficulty which is created by the fact that there is not, as is clearly shown in connexion with preventive inoculation against typhoid fever, any constancy in the immunising response of healthy men to one and the same dose of one and the same vaccine.

And greater than all is the difficulty which is created by the fact that there are very great differences in the effect produced by one and the same dose of vaccine in the infected as compared with the healthy, and in the severely infected as compared with the lightly infected. For this reason it is impossible to foresee accurately the effect which a vaccine will produce when inoculated into an untried patient.

Lastly, still further difficulties arise where we are asked to predict, instead of the result of an isolated inoculation, the effect of a whole series of inoculations periodically undertaken upon a patient. The difficulty of prediction here is insuperable, owing to the circumstance that, either as the result of the patient's personal factor, or as the result of the special features of his infection, the point at which he fails to respond to small immunising stimuli, and the point at which he tolerates and responds to larger immunising stimuli, are reached in the one case earlier and in the other case later.

While the fact that our powers of prevision are thus limited must be emphasised and kept in view, there would be grave error in supposing that we can never predict the results of an inoculation. If there is, in connexion with bacterial vaccines, one thing which is more assured

n another, it is that it must always be possible, after it may be a gthy process of trial and error, to arrive at a dose regarding which it l thenceforward be possible to predict that it will, when employed on patients who are suffering from a localised infection of strictly ited extent, produce either a very slight negative phase followed a positive phase, or a positive phase without the intervention of y negative phase. Such doses, when they have once been arrived at, y be safely employed in the treatment of strictly localised infections art from an exercise of control on the part of the practitioner.

It may be predicted, in connexion with a staphylococcus vaccine, at it will, when administered in a dose corresponding to 100 million staphylococci to a patient, who is developing an isolated furuncle, actically always produce an immediate positive phase and arrest the velopment of that furuncle. It may further be predicted of a dose rresponding to 250 to 300 millions of staphylococci, administered three four days later, that it will reinforce the action of the previous dose, d practically always put an end to the furuncle. Again, it is probably fe to predict of a streptococcus vaccine, which has been prepared from ordinary case of erysipelas, that it will, when inoculated in a dose rresponding to 2,000,000 of streptococci, abort, or at any rate tempor- ily arrest, an incipient streptococcic lymphangitis. In like manner, here we have to deal with a tubercular adenitis or arthritis of strictly nited extent, it may be predicted that an initial dose of 1–20,000 mgr. a dried and comminuted tubercle culture will give a satisfactory sitive phase without the intervention of any serious negative phase. , however, inquiry were made for a dose of the same vaccine, which ould apply to all tubercular patients without distinction, that demand ing coupled with the condition that a serious negative phase should not, any case, be produced, the dose would probably have to be fixed at t larger than 1–50,000 mgr.

The question of the appropriate dosage to employ, when taking in nd a case of tubercular infection carries us on—since here there can no question of eradicating the infection by one or two inoculations -to consider the question whether it would be possible to lay down scheme of dosage such as would comply, on the one hand, with the oviso that a positive phase should follow upon every inoculation d, on the other hand, with the proviso that no serious negative phase ould in any case result.

Assuming a case of strictly localised tubercular infection of limited tent, and postulating inoculations at 10-day intervals, it might perhaps tentatively laid down that, starting with a dose of 1–20,000th mgr. Koch's tubercle powder, this dose might, as a rule, be increased every onth by gradual increments up to 1–4,000th mgr., this latter dose eing reached only after the expiration of six months' treatment. There ould indubitably be many cases which would benefit under the exhibi- on of more generous doses. And, again, there would be other cases

where, after years of treatment, doses of 1–20,000 mgr. could not with advantage be exceeded.

I think it will have become clear, from the consideration of these data, that if, in connexion with tubercular infection, a hard-and-fast scheme of dosage were to be laid down, such as might safely be carried out in the absence of any method for readjusting the doses, it would —and what applies in connexion with tubercular infections applies *mutatis mutandis* to all other chronic infections—it would, I say, be necessary to employ almost minimal doses of vaccine. In point of fact, doses would have to be employed which would not allow of the attainment of either satisfactory or rapid results in the majority of patients. Some method of controlling and readjusting the dose to the special requirements of each individual case is thus seen to be essential.

Let me now turn to consider the suggestion that the clinical symptoms of the patient are capable of furnishing the guide of which the immunisator stands in need.

Suggestion that the clinical symptoms of the patient will furnish to the immunisator a guide by which he may regulate his immunisation procedures.

It is one thing to assert that it is very often possible for the immunisator to glean from the patient's symptoms information which would assist him in selecting his doses of vaccine—that such help can be obtained is a self-evident truth—it is quite another matter to put forward—as has recently been done—the suggestion which stands at the head of this subsection. We have it here implied that the clinical symptoms of the patient will in every case inform us whether we have been employing the proper quantum of vaccine, or too large, or too small a quantum. It is the suggestion thus understood which I propose here critically to consider.

If I were directly addressing the authors of that suggestion, I do not say that I might not be tempted to deal with it by the Socratic method. I might in such case, with a view of convicting its authors of having put forward an ill-considered proposal, press for answers to the following questions. I might quite well, addressing myself to them, ask :—

(*a*) Is it quite certain that there will, in every case where immunisation may be called for, be clinical symptoms to guide you in your dosage ?

(*b*) Given the case where the patient presents an obvious pathological condition (I have in view here either a definite constitutional disturbance or a definite local lesion), what will be the symptoms, which will tell you that you have inoculated, as the case may be, an appropriate dose, or an excessive dose, or too small a dose of your vaccine ; and, again, what will be the signs by which you will know that the hour has arrived for reinoculation ?

(*c*) Is there any certitude that such changes, as your inoculations

y be capable of producing, will, in every case, be produced with icient promptitude to inform you before the inoculation next in series s due, whether you ought to employ the same dose of vaccine, or increase diminish the dose ?

(*d*) Is it inconceivable, in the case where the localised focus of infection hut off from the blood stream, that you may obtain by inoculation effect upon the circulating blood without obtaining a corresponding nge in the condition of the focus of infection ?

(*e*) Or finally, recasting a verse from Kipling into a question—

> How do you *know* that your God will rouse you
> A little before the nuts work loose ?
> How do you *know* that *His* pity allows you
> To leave off from work whenever you choose ?

Instead of merely suggesting that these questions demand answers, will try to supply these. Let me begin by putting it to you that we ve, in connexion with the question as to whether we may trust to e clinical symptoms to serve as guides to us in therapeutic inoculation, consider four different classes of cases. We have :—

(1) *the case of a localised infection, which is of such a nature that the nical observer can immediately either see for himself or learn of every ange which occurs in its condition ;*

(2) *the case of a localised infection where the conditions are unfavourable the observation of changes in its condition ;*

(3) *the case of an acute febrile condition ;*

(4) *the case where all local and general symptoms are in abeyance.*

Let me deal with each of these cases seriatim, and before we have one, it will be found that an answer has been furnished to each of the uestions I have formulated above.

(1) *The case of a localised infection, which is of such a nature that the inical observer can immediately either see for himself or learn of every ange which occurs in the focus of infection.*

The most typical examples of such infections as these are the staphy-ococcic infections of the skin and subcutaneous tissues, to wit, furuncu-osis, suppurating acne, and sycosis. These are representative, not nly in the respect that they are directly accessible to inspection and alpation, but also in the respect that they may rapidly become better r worse. This acute evolution is, as consideration will show, very aterial, inasmuch as we can in such cases count on being put into ossession of the results of the foregoing inoculation before we are called pon to undertake the inoculation next in series. In the same class vith these may be placed the streptococcal infections of the skin and ubcutaneous tissue which take the form of impetigo, "serous furuncles," rysipelas, and lymphangitis.

In the category of localised infections whose evolution can be readily ollowed, may further be placed infections of wound surfaces and mucous

membranes by pyogenic micro-organisms. Here information can b drawn, not only from the appearances of the suppurating surfaces, bu also from the amount of discharge. In the case where the seat of th infection, though communicating with the exterior, is more deep-seate —as, for instance, in the ear or uterus or male urethra—we can stil draw certain inferences from the amount of the discharge and fron its character.

In the category of local infections, whose course can be readily fol lowed, may be included also many of the localised infections with whicl we have to deal in connexion with the kidney, the bladder, the colon and other organs. Here we may glean information with regard to th condition of the focus of infection by noting the character of the excretions or, as the case may be, the amount of the secretion. We can, for instance measure the quantity of albumen and the amount of pus in the urine We can, again, in the case where we are immunising against a coli infectior of the urinary tract, watch for that characteristically rapid clarificatior of the urine by flocculation and sedimentation which comes under observa tion when the microbes undergo agglutination in the urine. In colitis we can watch for a change in the excretion of mucus and in the shedding of the membranous casts. In Miculicz' disease—a disease which appears to be generally associated with a streptococcus infection of the salivary and lachrymal glands—we can take note of the variation in the amount of saliva and tears. And in those cases of cholecystitis, where, after operation, the bile evacuates itself through the operation wound, we can similarly obtain some information with respect to the infection by noting the changes in the bile.

Where the localised focus of infection is so situated as to interfere with a delicate reflex mechanism—I have in view here the case where frequency of micturition results from a bacterial infection—we have, again, a sensitive recording mechanism which may keep us informed with respect to the progress or regress of the infection.

And these objective signs, in connexion with the focus of infection, are not our only means of learning of the condition of the local focus. Where we have, in association with a local focus of infection, pain or discomfort, and, of course, we have these in connexion with very many forms of localised infection, we can very often, through the intermediary of these, learn of any changes which may occur in the focus of infection. It will thus be seen that, by one way or another, we may, in connexion with a large number of localised infections, hope to be kept informed with respect to their progress by the intermediary of the clinical symptoms.

The problem as to how we are to interpret this information and to turn it to account in regulating our dosage is very far indeed from being a simple problem. We have, however, in our hands an important clue to the interpretation of the clinical data when we call to mind that—

(*a*) A moderate dose of vaccine, such as would generally be the most

ful dose in connexion with a chronic infection, produces, after a comatively short negative phase, a positive phase which might, on an rage, extend over the interval between two successive inoculations lertaken some 10 days apart.

(*b*) A smaller dose would produce an immediate positive phase, but ositive phase which would not be maintained for more than a very days.

(*c*) An excessive dose would produce a negative phase, which would through a large part, if not through the whole, of the interval which ally elapses between two inoculations.

Keeping the graphic curves, which correspond to these three types response, clearly before the mind's eye, we can now take the clinical tory and see which of these different types of curve it will fit in with.

The chronology of the different incidents will here give most important ications. Where, in connexion with pustular acne or furunculosis, re has been a fresh outcrop of spots, or a new furuncle, or increased stulation, or where, in connexion with tubercular cystitis, there has n more pain and increased frequency of micturition, or where, in nexion with an infected wound or mucous membrane, there has been re discharge, or where, in connexion with a gonococcal or tubercular hritis or in connexion with a tubercular adenitis, previously painless, n has developed after inoculation, we may take it that there has been egative phase, and that the type of response is that which is obtained the inoculation of a moderate or, as the case may be, an excessive se. Where the negative phase symptoms have been severe or proged it will be plain that the dose of vaccine has been excessive.

Where, on the contrary, we learn that there was improvement for ew days after inoculation, and afterwards a relapse to the conditions ich obtained before inoculation, we may suspect that our dose of ccine has been too small.

These conclusions will be confirmed if there has been associated with e exacerbation of the local symptoms, a rise of temperature, or a general eling of malaise and constitutional disturbance, and, with the improveent in the local symptoms, a condition of euphoria.

Fallacies in connexion with the interpretation of the clinical symptoms.

It might be supposed that, with the above clue in our hands, the terpretation of the clinical symptoms would, at any rate, in the class cases we are here discussing, give us quite adequate guidance in the atter of dosage and the interspacing of our doses. This is very far deed from being the case.

On the one hand, the conditions in the focus of infection may upon casion be quite fallacious guides. And, on the other hand, the conitutional symptoms cannot be relied upon to convey all the facts which e material for us to know.

These points with regard to the fallaciousness and incompleteness the clinical record, even at its best, may be conveniently considered

here. We then need not come back upon them when we come to deal wit the three other categories of clinical cases which were enumerated abov

(a) *The clinical symptoms which are associated with the focus of infectio may convey quite an erroneous idea of the conditions which there obtai*

Where, as is so often the case, a lupus which is infected by strepto cocci is being treated with inoculations of tubercle vaccine only, th local appearances may be very fallacious. In such a case progress i the direction of the suppression of the tubercular infection may quit well be masked by the persistence, or, as the case may be, incident exacerbation of the streptococcic infection. Where, in a case of com bined infection by two species of microbes, both the microbes are attacke by vaccine-therapy, with the result that one is successfully combate while no headway is made against the other, the appearances may quit well suggest to the purely clinical observer quite an erroneous idea o the state of the case.

Where we are dealing with deep-seated infection we may erroneousl attribute significance to an increase or to a recurrence of œdema. I illustration of the opportunity for error, which presents itself in connexio with œdema, I may instance the case of a patient who had bee discharged as cured of a lupus of the nose, and who, alarmed at a recurrenc of the swelling and reddening, recently came back again for furthe inoculation treatment. On examination of this patient's opsonic inde a normal reading was obtained. It was also noticed that the patient' fingers were badly chilblained. This fact, taken together with th circumstance that her blood coagulability was markedly diminished suggested that we had before us not a recurrence of the lupus, but a chilblain on the nose. This inference was confirmed when the swellin and reddening of the nose, and in association with this the chilblain on the fingers, rapidly disappeared under the influence of calcium lactate.

It may quite well be that variations in the size of tubercular gland and perhaps changes in the amount and effusion in tubercular joints may in similar manner occur, independently of any progress or regress of the infection, directly as the result of changes in the coagulability and viscidity of the blood.

(b) *The clinical symptoms in connexion with the focus of infection even when they convey accurate information with respect to the conditions which there obtain, may suggest to us quite a wrong idea of the conditions which obtain in the circulating blood.*

I have often pointed out that we have no right to assume from the fact that the conditions in a patient's blood are unfavourable to the growth of microbes that we have conditions unfavourable to their growth also in every other region of the body. Where the conditions in a nidus of bacterial infection have been investigated, it has almost invariably

[1] *Vide* author's paper: "On the Pathology and Treatment of Chilblains," *Lancet*, Jan. 30, 1897. *It may be incidentally noted here that chilblains, occurring in an adult who has not suffered in this way in childhood, and occurring in association with indolent nodes in the hands, often give indication of the presence of tubercular infection.*

ıed out that these have been much more unfavourable to the destruc- ı of micro-organisms than those in the blood.

Into the reasons of this I propose, in the second part of this paper, ;o more fully. For the present it will suffice to point out that it follows ·ctly from the fact of the conditions in the blood differing from those he nidus of infection, that the local symptoms cannot—even in case y convey to us an accurate account of the conditions of the microbial ·ction in the local focus—give us a true measure of the conditions ch obtain in the circulating blood.

The significance of this conclusion cannot escape you. You will reciate that, inasmuch as the object of all inoculations is to operate the condition of the blood, and inasmuch as the success or ill-success inoculations can be judged only by the effect which is exerted on blood, and inasmuch as the symptoms which manifest themselves he focus do not furnish an unfallacious index of the condition in the ·ulating blood, we have not in those symptoms any unfallacious de for control of the dosage.

(c) *The fact that the patient's general clinical condition remains undis- bed does not warrant us in assuming that the antibacterial potency of blood is not undergoing momentous fluctuations under the influence spontaneous auto-inoculations.*

We have just seen that the observation of the focus of infection ·apable of suggesting to us quite a false idea of the effect that has n produced upon the blood by inoculation. Equally important is or us to appreciate that we may be led quite as far astray if we take ıpon ourselves to infer, from the fact that the patient has remained e from constitutional disturbance, that his blood has not, under the luence of spontaneous auto-inoculations, undergone alterations such as immunisator ought to take into account in regulating his dosage l in interspacing his inoculations. Again, we should fall into error if were to assume of every slight constitutional disturbance that it must nd in connexion with an auto-inoculation. From the responsibility guessing in such a case, and from the dangers of guessing wrong, there ıuld seem to be no way of escape except that which is provided by ood examinations.

Thus far I have spoken only of the case of a localised infection which es to the clinical observer notification of every change which occurs its condition. We have next to consider the case where changes in e focus of infection do not manifest themselves either rapidly or clearly on the clinical record.

(2) *The case of a strictly localised infection where the conditions are favourable to the observation of changes in the condition of the focus infection.*

Typical examples of the class of local infections I have here in view e furnished by the majority of cases of tubercular adenitis, tubercular thritis, lupus, and pulmonary phthisis. Here we are dealing with

processes which are so slow in their evolution that one cannot, between one day and the next, or between one week and the next, or even it may be between one month and the next, make certain whether there has been progress or regress.

It is, of course, obvious that the clinical symptoms in connexion with the focus of infection will ultimately tell us whether vaccine-therapy has been of benefit, or has been productive of harm, or, as the case may be, that it has effected no sensible change in the focus of infection. But information which arrives thus tardily, arrives too long after the event to be of service. It is from the point of view of its unfruitfulness to the patient to be classed almost in the same category as the retrospective information which the physician derives from his post-mortem examinations and his case-mortality statistics.

For where we discover, only after a long sequence of inoculations, that the scheme of dosage which we adopted was appropriate to the conditions of the case, we have no certainty that the situation has remained the same and that the same scheme of dosage is still appropriate. The verdict of the clinical symptoms on that point will, if the inoculations are continued, be furnished only after another period of months, and then the problem will once more present itself and the answer will once more be postponed, and so on indefinitely.

Again, where we discover, after a long sequence of inoculations, that sensible harm has been done, we learn only that our scheme of dosage has been ill-chosen, but we do not know in what direction to seek for a better scheme. In like manner where, after a long sequence of inoculations, we discover that the condition of the focus is unaltered, we do not know whether some of our sequence of inoculations have been benefiting the patient, or whether others may not have been doing him harm, or whether they have all been alike ineffective.

The German proverb speaks here truth when it tells us that "God Almighty does not send in the reckoning on every Saturday night." That reckoning comes, in cases such as we are here considering, only after long waiting. And when it comes, it comes not in the form of a detailed account with each item set out with its profit and loss, but only as a grand total. It is therefore the part of a prudent man anticipating the day of reckoning to make periodic inquisition and to find out for himself how his account stands.

(3) *The case of an acute febrile condition.*

In the respect that we are here face to face with a pathological phenomenon, whose course can be continuously followed by the clinician, the case of an acute febrile condition presents analogies with the case of a localised infection which has an acute evolution, and which is directly accessible to inspection. In fever we have, in the readings of the clinical thermometer, a means of judging of the results of our inoculations which is of equal value with the indications which are, in the cases of localised infection, afforded to us by a direct inspection of the focus.

While the temperature curve is a measure of intoxication and not of unisation, and while there is no direct and constant relation between temperature curve and the production of antibacterial substances ie organism, we may take it that a diminution in the antibacterial ncy of the blood generally leads to a multiplication of microbes ie system, this to increased intoxication, and this, in its turn, to a of temperature. In like manner, we may take it, that an increase ie antibacterial potency of the blood leads, as a rule, to a restriction icrobial growth in the organism, this to diminished intoxication, and in its turn, to a reduction in the temperature. But while it ie rule to find this inverse relation of temperature to antibacterial ncy, it is certainly not the invariable rule. It is notorious that ssive intoxication may condition a fall in temperature; and it is eivable that a rise in temperature may sometimes be directly associ- with efficient immunising response.

We must, as in connexion with localised infections so also in connexion pyrexia, be on our guard against the fallacy of mixed infection. as the aggravation or persistence of clinical symptoms in connexion a local focus may sometimes be imputable to an intercurrent, or ady pre-existing but subordinate infection, so here, in connexion pyrexia, a rise or fall of the temperature may sometimes be imputable bacterial infection other than the one which is being combated.

4) *The case where all local and general symptoms are in abeyance or returned to the condition which prevailed previous to inoculation.*

The case I have here in view may be encountered indifferently in nexion with the three classes of infections considered above.

In connexion with the first class of infection, and let us here take the of a furuncular infection, the situation arises when, after the ppearance of all active furuncles, a condition of uncertainty pre- s as to whether the patient is now immune, or whether he is still le to suffer at any moment from a recurrence of his troubles.

In connexion with the second class of cases, let us say, in the case of berculous joint, the problem presents itself where the clinical symp- s have quieted down, and we are left in a state of uncertainty whether infection has been eradicated or whether it still persists.

In connexion with a pyrexial infection, we are confronted with the culty when we are called upon to decide whether the invading microbes e been eradicated or whether there is still liability to recurrence.

It will be manifest that in none of these situations—and let us note that ations such as these must sooner or later arise in connexion with every nunisation which is progressing towards recovery—can the immunisator ain any guidance or help from clinical methods.

Résumé of the conclusions which have been arrived at.

Let me now try to sum up to you very briefly the main conclusions ch have been reached in our inquiry as to how far, and in what kind

of case, the clinical symptoms enable us to correct and control the dosage of our vaccines.

Pro hac vice, I may perhaps with advantage discard the vocabulary of science for the vocabulary of metaphor.

(1) Where it is a question only of a short passage, or, as the case may be, of a succession of short passages, with landmarks continuously in view, over familiar waters where, while stranding is not out of question, serious shipwreck is unthinkable, it would be ridiculous to insist that the pilot should keep the lead going, and should set his course by compass readings. On the other hand, even here, if, as often happens, the landscape becomes indistinct, or is temporarily obliterated, the only rational alternative to anchoring and waiting for the reappearance of the landmarks would be to proceed by compass readings and by the lead.

(2) Where it is a question of a protracted voyage, during which no sequence of guiding landmarks could be expected to come into sight, it is desirable that compass readings should be taken every time that the ship is put upon a new course, or, if that should be impossible, always at the very first warning of impending danger. For otherwise the vessel might very easily zigzag about to no purpose, or she might, if the error in shaping her course happened to be a cumulative one, go very far out of the right track and come to serious disaster.

And we must here beware of thinking that the possibility of error in connexion with the laying off of the ship's course is the only possibility of error which we have to take into consideration. There are cases in which the ship may be flung violently off her course by the buffeting of winds and seas. And there are cases where, under some bias of wind or current, the ship may come off her course in a perfectly insensible manner. She may then easily run on the rocks if we do not lay out our bearings by the help of the compass.

(3) Where it is a question of navigating through specially perilous and uncharted waters, where a wrong turn of the helm might at any moment bring disaster, he would be a very imprudent and reckless seaman who would dispense with the aid of lead or compass, even when there were landmarks in sight. Continuous soundings might here quite well give us timely warning of dangers, of which, even with a good lookout, we might be unaware until we had already run in amongst them. Nor would the prudent seaman discard compass and lead until the vessel had come safely into harbour, or until, as the case might be, all efforts to save the ship had been abandoned.

My metaphor will have accomplished its purpose if it has brought home to you that, while we can in many cases conduct our immunisations aright by relying on the experience gained in the past, and upon clinical observation, there are whole classes of cases—these being cases where the only hope for the patient is to be found in the proper conduct of immunisation procedures—where the clinical symptoms cannot be trusted to furnish the necessary guidance.

If this is so, it will be well for us to consider how it has come about t the clinical symptoms have been proclaimed to be an all-sufficient de. I suggest that the explanation of this is to be found, not in any ıbt as to the reality of the risks which might be incurred by the adıistration of excessive doses of vaccine, but in the persuasion that, if y the glaring errors which were committed in connexion with the ginal inoculations of Koch's tuberculin are avoided, the results of cine-therapy will be, on the whole, satisfactory. Associated with s is, I submit, also the further persuasion, that where failures occur responsibility for these need not necessarily rest with the immunior, and that such responsibility can at any rate never be brought home him.

I see nothing to censure in this attitude. It is clearly an attitude ich conforms in every point to the accepted code of medical ethics. t, at the same time, I cannot conceal from myself that in every other partment of practical life—let us think, for instance, of shipping or lways or any engineering industry—a much higher standard of reonsibility is enforced. It is only in connexion with medicine that the pert is content to aim at an average of satisfactory results ; it is only medicine that he thinks efforts to secure uniform success uncalled for ; d it is only in medicine that he contends that he has at disposal all the ormation he requires for his guidance, when a moment's consideration uld make it clear to him that he is taking risks, and is proceeding in e dark.

Of a certainty this will not go on for ever. The higher standard of reonsibility, which is enforced everywhere else in civilised life, will ultiately be enforced also in medicine. It will be enforced in medicine when shall have shaped it into something more like a scientific profession.

We have here come, as you will immediately perceive, face to face th the critical question as to whether vaccine-therapy has been placed on a definite scientific footing by the technique, which, in conjunction th Leishman, Douglas, and others of my fellow-workers, I have elaboted for measuring the changes in the blood which follow upon inoculaon. I refer here in particular, but not exclusively, to the technique r the determination of the opsonic index of the blood.

ɔes the technique for the determination of the opsonic index furnish to the immunisator accurate and useful information with respect to the way in which the patient is progressing ?

It will not have escaped attention that while the statements, which my llow-workers and I have made with respect to the accuracy of the opsonic chnique and the diagnostic value of the opsonic index, have been lly confirmed by a large body of workers, the technique has, by certain ithors, been denounced as flagrantly inaccurate, and it has been asserted nfidently, and with an imposing show of figures, that there is no differ-

ence between the serum of the infected and the uninfected man in th matter of its power of inciting phagocytosis.

The proper reply to such assertions is to set out a large number o duplicate experiments and counts showing the consistent results whic the technique yields in the hands of competent workers, and then t leave it to the authors, who have obtained only inconsistent result to explain whether their results are to be ascribed to a want of intelligen appreciation of the principles of the technique, or to an incapacity fo that conscientious attention to detail which the technique exacts, or t a bias which leads to the scamping of the precautions which ar required for the avoidance of error.

My friend and fellow-worker, Dr. Alexander Fleming, has been goo enough to come to my aid in this matter, and to extract from our labora tory records a large number of control experiments which were mad in the course of our ordinary work. He has supplemented these by carry ing out, both independently and in association with others of our labora tory workers, a large number of further control experiments. Dr Fleming has also taken upon himself the labour of investigating, in methodical manner, the effect of certain errors in technique, and, i particular, the effect of a fallacy in connexion with the agglutinatio of red blood corpuscles which has been only recently encountered by us The results of these inquiries will be found set out in a paper which appear in this number of *The Practitioner*.[1]

The results,[2] which are there placed on record, demonstrate in a con

[1] *Practitioner*, May, 1908, pp. 607–634.

[2] While these, which concern the only vital points in connexion with the tech nique, are fully set forth in Dr. Fleming's paper, there are two further points i connexion with the measurement of the opsonic index which are reserved for furthe study. The *former* of these relates to the question as to whether trustworthy result can be obtained by the opsonic technique in cases where the patient is suffering fro severe bacterial intoxication. The *second* question as to whether estimations of th opsonic index made in different laboratories ought, given accurate work, to show th same close agreement as duplicate estimations made in the same laboratory.

Question as to whether trustworthy results can be obtained by the opsonic techniqu in cases where the patient is suffering from a severe bacterial intoxication.

A priori considerations would suggest that, in cases of severe bacterial intoxica tion—and *mutatis mutandis* the same applies to "absorption experiments" made t test the specificity of the opsonins of normal blood—there may quite well com into consideration in addition to such effect as may be exerted upon the bacteri by the opsonins of the serum also a toxic effect exerted upon the leucocytes by th bacterial toxins in the serum. At any rate, whether this is the explanation o the matter, or whether that explanation is to be sought in the non-exclusion o the fallacy of hæmagglutination which is wont to occur in such cases, it is to be ob served that, in two series of experiments, which were undertaken in our laborator collectively by the staff and by some workers who were attending the course o instruction, very inconsistent results were obtained with the bloods derived fro two patients who were suffering from severe bacterial intoxication, while the result obtained in the case of the control bloods showed in both cases almost ideal agree ment.

Question as to whether estimations of the opsonic index made with different culture in different laboratories ought, given accurate technique and accurate work, to sho the same closely concordant results as may properly be exacted in the cases of duplicat estimations made with the same cultures in the same laboratory.

While it is of course well understood that where an opsonic index is to be deter

ve manner that in connexion with tubercle—and it is only in con-
on with tubercle that the accuracy of the method has been specifically
d in question—the opsonic technique gives, in competent hands,
emely consistent results, and that it gives these, not only in the case
ormal uninfected persons, but also in the case of the infected persons
whom we have dealt in our hospital.

[need not say any more but pass on to consider the utility of the rmation furnished by the opsonic index. This may be considered er two headings. We may consider first what has been learned in way of principles and facts of general application by the measurements he opsonic index, and secondly what help is furnished by estimations he opsonic index in connexion with the treatment of a case.

A few words will suffice in connexion with each of these questions. onnexion with the former, I would draw your attention to the fact —if we except the case of immunisation against typhoid where the nges in the bactericidal and agglutinating powers of the blood have ed as a guide—almost everything that we know of the laws which ern the immunising responses which are evoked by bacterial vaccines nan, everything that we know of the phenomena of autoinoculation, everything that we know with respect to the proper dosage of our cines, is derived from the opsonic index.

If any light has been thrown on the machinery of immunisation in , if it has been possible to substitute for the excessive doses of erculin which were originally employed, as a first step, the imal doses which could be administered without injury, and after- ds, both in connexion with tuberculin and in connexion with a num-

d, exactly comparable quantities of serum, exactly comparable quantities of ocytes and exactly comparable numbers of bacteria must be employed in the s which correspond respectively with the patient's blood and the normal blood, is a further theoretical requirement which had been overlooked. Where icate pairs of experiments, undertaken with different bacterial suspensions, o give concordant results, the microbes employed must in each case be com- ble from the point of view of their power of resisting phagocytic attack. Now re not entitled to assume of all the tubercle cultures, which might come into ication in different laboratories, that they correspond in this respect; and it is *iori* probable that this property may be modified to a varying extent by the rent treatments to which the cultures may be subjected in different laboratories. le this possibility must always be kept in view, it would appear, from such con- experiments as we have carried out, that the mode of culture and the mode of aration does not in the case of the tubercle bacillus make any important differ- , if indeed it makes any, in the opsonic index arrived at.

t is quite otherwise in the case of certain other microbes, in particular in the of the bacillus coli and the meningococcus. Here (and this has already been ted out by Houston in connexion with the meningococcus) very different results be obtained when bloods are tested with different cultures all derived from same stock. For in association with the attenuation which these microbes rgo upon artificial media, they gradually become less and less resistant to the ocytic attack of the normal blood. A comparatively low index is now obtained e before a very high index was obtained, the result being of course simply due increase in the denominator in the fraction:

$$\frac{\textit{phagocytic count of the patient's blood}}{\textit{phagocytic count of the normal blood.}}$$

ber of other bacterial vaccines, the minimal doses which can be employe with effect, and if we are now in a position to interspace our doses o vaccines in connexion with scientific principles—all this is due to th technique of the opsonic index.

When we turn from the general to the particular and ask what hel we can gain from the determination of the opsonic index in connexio with the treatment of individual cases, I would point out that we can by the aid of the opsonic technique, determine for each individual cas exactly the same points, in connexion with the dosage, as have alread been determined by the technique for the average case.

Knowing as I do full well that we have not in the opsonic reading anything even remotely comparable with the sextant and chronomet observations, patent log readings, and enumerated revolutions of th screws by which the position of an Atlantic liner is fixed from hour t hour as she crosses the ocean, I was, as you will perhaps have remarke careful when employing the language of metaphor to speak of th measurement of the content of the blood in protective substances a analogous to the soundings and compass bearings of the navigator rath than to the more highly developed methods of nautical science.

While the modern navigator is in a position, in most cases, to prick o his position upon the chart with accuracy, the immunisator of to-day less fortunate because less well equipped with scientific methods, can a least tell by his measurements of the opsonic power, or, as the case ma be, by his measurement of any of the other protective elements of th blood, whether he is shaping a proper course for his patient, and wheth his patient is keeping on that course.

Let me now pass to consider the second division of my subject-matte

PART II.

How may such antibacterial agencies as the patient may already possessed of or may acquire by immunisation, be directed to t destruction of microbes in the focus of infection?

Where inoculations are undertaken for prophylactic purposes, t immunisator is not required to make any dispositions for bringing t antibacterial elements of the blood fluids and the phagocytes into appl cation upon invading microbes. They will come into application in perfectly automatic manner.

Exactly the same thing holds true in connexion with vaccine-thera where the invading bacteria have not as yet effected any fundamen alterations in the invaded tissues.

Entirely different is the situation when vaccine-therapy is resorted

ises where the invading bacteria have already firmly ensconced
selves, and have profoundly modified the conditions in the focus of
tion. Here it will not suffice to increase and maintain the antibac-
l power of the blood. It will be necessary in each case to take special
to bring the phagocytes and antibacterial elements of the blood
s to bear upon the invading micro-organisms.

Before we can profitably consider what these steps ought, in each
cular case, to be, it will be well to take a general survey of the
itions with which we have to deal.

Ve may study, *first*, the distribution of the antibacterial agents in
normal organism; *secondly*, the inflammatory reaction which super-
s upon a bacterial invasion of the tissues, and the changes in the
ibution of the antibacterial agent which are associated with that
ion; and, *thirdly*, the conditions which present themselves when
obes have survived the inflammatory reaction, and have made for
nselves a nidus in the tissues.

eral question of the distribution of antibacterial agents in the normal organism.

In the circulating blood the organism has at immediate disposal
tically the whole of its defensive forces. It has here at disposal,
he one hand, the bacteriotropic substances which are contained in
plasma and, on the other hand, its whole force of phagocytes.

It follows that where microbes make their way into the blood stream
e will come into application against them, not only a superabundance
hagocytes, but also a superabundance of bacteriotropic substances.
se last will operate upon the bacteria with a mass effect which will be
inuously kept up to a level corresponding with the full bacteriotropic
sure of the patient's blood.

Where, on the contrary, the microbes effect an entrance into the
ues, they will find opposed to them, for the moment, only such stray
gocytes as may be casually passing through the tissue spaces which
the subject of invasion, and only such quantum of antibacterial sub-
ces as may be contained in the lymph which is flowing through these
icular tissue spaces.

It is, in view of these considerations, immediately intelligible that the
nal organism should, as it does, successfully resist microbes, admin-
ed through the channel of the blood, when it fails to offer resistance
he self-same microbes, administered by way of the tissues. It is
in conformity with these considerations that localised infections
ld be ordinary every-day events, that these localised infections should
very rarely lead on to secondary septicaemia, and that primary
icaemic infections should be of comparatively rare occurrence.

From these general considerations, we may also draw the lesson that,
ll cases of localised infection, the aim and object of our treatment

ought to be to level up the conditions in the infected tissue to the conditi in the circulating blood.

Nature of the inflammatory reaction which supervenes upon a bacte invasion of the tissues and changes in the distribution of the antib terial agents which are effected by that reaction.

No one who has perused the illuminating lectures in which Metchnik discourses on the Comparative Pathology of Inflammation will fail to c to mind his great generalisation that everywhere through the invertebr and vertebrate kingdoms the intrusion of bacteria or other foreign e ments into the tissues is responded to by a determination of phagocy to the seat of injury. Nor will any one who has read those lectu have forgotten that Metchnikoff there contends that the phagocy are the only antibacterial agency, and that in the emigration of phagocy is to be found the whole import of inflammation.

Since the publication of the lectures, we have in connexion wi immunity been moving on to new view-points. Much has since th been learned about the antibacterial powers of the blood fluids, and particular the work of Douglas and myself has shown that phagocyt when unaided by the opsonic power of the blood, are comparative impotent. It has accordingly become impossible to confine our atte tion, as did Metchnikoff in his lectures, to the phagocytes, and to rega these as alone significant, overlooking the increased transudation fluid from the blood vessels which invariably accompanies inflamm tion, or setting this aside, as Metchnikoff did, as a phenomenon which no special significance was to be attributed.

In short, we can no longer see in the transfer of phagocytes from t circulating blood to the invaded tissues the whole import of inflammatio Rather we must to-day recognise in inflammation a process which mi isters to immunisation, on the one hand, by the transfer of phagocyte and, on the other hand, by the transfer of antibacterial fluid from t circulating blood to the invaded tissues.

When we correlate with this view of inflammation the therapeutic principle which was borne in upon us by the consideration of the distr bution of the antibacterial agencies in the normal body, it becomes clea that the therapeutical principle in question—I mean the principle levelling up so far as may be the conditions in the invaded tissues to thos in the circulating blood stream—is neither more nor less than a polic of bringing intelligent aid to nature, and furthering the defensive measur by which the organism responds to bacterial invasion.

Moreover, it will presently appear that while the original theory Metchnikoff failed to furnish any explanation of the frequent ineffe tiveness of the inflammatory reaction, the theory of Metchnikoff as her amended puts into our hands a key which unlocks the problem of th frequent miscarriage of the inflammatory reaction, supplying at the sam time indications as to how such miscarriage may be rectified.

nditions which present themselves in the case where microbes have survived the inflammatory reaction, which has supervened upon their invasion, and have established themselves in a nidus in the tissues.

A nidus, in the sense in which I here employ the term, is constituted ıenever, as the result of the miscarriage of an inflammatory reaction, .d of changes in the tissues and products of inflammation, conditions e established in the focus of infection which are specially favourable the infecting microbes.

Common to every bacterial nidus is the circumstance that the effective cess of phagocytes and bacteriotropic substances to the infecting microbes in some way hindered.

The impediment may consist in any of the following:—(*a*) a ∙fective blood supply to the seat of infection; (*b*) a hypercoagu-ble and hyperviscid condition of the blood, for this would impede ansudation of fluid from the blood vessels and would be favourable to ıe coagulation of effused lymph in the tissue spaces; (*c*) an accumu-tion of excessive fluid in the focus, for this would prevent the phagocytes ɔming in contact with the bacteria; (*d*) a blocking of the tissue spaces y accumulated leucocytes and coagulated lymph, for this would constitute mechanical hindrance to the entrance of fluids and phagocytes into the ɔcus of infection; (*e*) a stagnation of lymph in the focus of infection, ɔr this would lead to a gradual reduction of the antibacterial potency f the lymph, which would render the phagocytes ineffective, and an ac-umulation of bacterial toxins and tryptic ferment, which would paralyse ıe phagocytes.

When we have appreciated that inflammation is associated with a ·ansudation of blood fluids and an emigration of phagocytes, and when e have appreciated that inflammation may be associated with hyper-lasic changes in the tissues, and may be complicated by secondary changes ı the products of inflammation, and when we realise that one or more of ıese factors may predominate over the others, it will inevitably come ome to us that we must expect to meet a number of different types of idus, each corresponding with an inflammatory reaction which has ıiscarried in some special way, and each possessing characters which re special to itself.

The more important cases with which we have to deal are the following:

(*a*) the case where serous effusion is the characteristic feature in onnexion with a nidus of infection;

(*b*) the case where the tissue spaces in the nidus of infection are blocked vith leucocytes and coagulated lymph;

(*c*) the case where suppuration has occurred in the nidus of infection, ınd an abscess sac has been formed;

(*d*) the case where in connexion with a microbic infection of the kin a nidus has been formed under the shelter of a scab;

(e) the case where a nidus of infection has been formed on the surfa of granulation tissue.

We may consider these cases seriatim.

(a) *Conditions which obtain where serous Effusion is the Characteris Feature in connexion with a Nidus of Infection.*—In connexion with sero effusion, it is important to call to mind that, if we except the typho bacillus and the cholera vibrio, all the other pathogenic germs which con into consideration in connexion with human pathology have, so far they have been examined, proved completely resistant to the bactericid action of the blood fluids. It is an obvious corollary to this that ar inflammatory reaction, which begins and ends in a serous effusion, mu practically always be an ineffectual reaction. Furthermore, it wi be plain to consideration that where serous effusion predominates ove phagocytic reaction to such an extent as to make it difficult for the pha gocytes to find and possess themselves of the bacteria, the excess of effu sion must constitute a hindrance to the destruction of these by the agenc of the phagocytes.

There is yet a further point which has to be borne in mind in connexio with serous effusion. It is now a familiar matter that when microbe are brought in contact with the blood fluids they rapidly absorb fron these the bacteriotropic elements for which they have a chemical affinity It is a corollary to this that wherever within the organism a serous effu sion stands in contact with bacteria, it will lose more and more of it antibacterial potency. Douglas and I have shown that it actuall does so.

(b) *Conditions which obtain where the Tissue Spaces in the Nidus o Infection are blocked with Leucocytes and with Coagulated Lymph.*—Th conditions, which we have here to consider, may be regarded as th direct converse of the conditions which are met with in connexion witl serous effusion. Whereas *there* we had a preponderance of transudatior over emigration, *here* we have a preponderance of emigratior over transudation; and, whereas *there* the effused lymph had littl tendency to clot, *here* the effused lymph clots in the tissues binding th inflammatory products together into a consolidated mass.

Inflammatory reactions which eventuate in this manner may be met with both in animals and man. In the rabbit, for instance, pus would appear normally to take the form of a solid cheesy mass, consisting of leucocytes held together by meshes of fibrin. In man the inflammatory products take this form only under quite special conditions. They do so in acute infections of the lungs, perhaps most characteristically in croupous pneumonia. There, as you know, we have in the alveoli of the consolidated lung not fluid pus, but a solid inflammatory product consisting principally of polynuclear leucocytes embedded in a meshwork of fibrin. A quite similar fibrinous pus may be formed also in the spaces of the sub-cutaneous tissue. Where we speak of a carbuncle, or of brawny infiltra-tion, or simply of induration in connexion with any pyogenic invasion,

e are in reality speaking of lesions which are characterised by a more or ss complete infiltration of the tissue spaces by a fibrinous pus.

If we now ask ourselves in what manner the blocking of the tissue paces by a consolidated pus will influence the survival of the invading acteria, the answer immediately suggests itself. Such blocking of he tissues will prevent, or render difficult, the conveyance of any additional lymph or of any reinforcing phagocytes into the focus of infection. f now the phagocytes which are on the spot have, owing to deficient psonic power in the lymph originally effused, been from the outset ineffective; or, if having originally been effective, they have been confined y the coagulation of the lymph, or have been rendered ineffective, ither directly by the accumulation of bacterial toxins, or indirectly by ymph gradually losing its opsonic potency; it will be intelligible that he invading bacteria should—in the interior of the infiltrated tissues—e able to maintain themselves safe from attack, at any rate, until such me as the barrier against the protective agencies of the organism shall ave been broken down by the sloughing of the infected tissues.

(*c*) *Conditions which obtain in the case where Suppuration has occurred nd an Abscess Sac has been formed.*—What occurs in connexion with uppuration and abscess formation would seem to be as follows:—

When the leucocytes disintegrate in the focus of infection—and hey will die and disintegrate not only under the influence of the bacterial oxins which are generated, but also independently of this—there will, s has been shown in a masterly manner by Opie, be liberated in the focus f infection larger and larger quantities of tryptic ferment, until finally he antitryptic power,[1] which the lymph possessed when it transuded

[1] It may be of interest here to briefly describe a simple technique by which the ryptic potency of the leucocyte when isolated from the blood fluids, and the antiryptic power of those fluids may be demonstrated and measured. For the demonration of the tryptic power of the leucocyte we may have recourse either to leuocytes isolated from pus, or to leucocytes isolated from the circulating blood. Vhere we are concerned to avoid sources of error from bacterial contamination and o employ irreproachable leucocytes, we may most conveniently have recourse to he circulating blood. The procedure which will be appropriate will be indicated y the following instructions:—

Take a piece of glass tubing, about three-fifths of an inch diameter and 2 to ½ inches long. Draw it out at one end into a capillary stem forming thus a pipette vith a moderately long barrel-shaped upper extremity. Employing a rubber teat nd placing a mark upon the stem, draw up first any convenient measured quantity f blood (say, 50 to 100 cmm.) into the pipette, and then 5 volumes of distilled water. Iix in the barrel of the pipette and then seal off the capillary stem a little below the oint where it expands into the neck, and place the diluted hæmolysed blood, which vill thus have been obtained, in an incubator at 37 C. Leave it there for 30 minutes. Iow, using another capillary pipette, direct a stream of water on to the clot which vill have formed and continue washing the clot and removing the wash water until contracted and almost colourless clot shall have been obtained. (Microscopic xamination of such a clot shows that it consists of a solid mass of leucocytes embedded in a meshwork of fibrin.) Now liquefy some 10 to 15 per cent. gelatine—a ube of the nutrient gelatine which is employed as a culture medium will serve for this urpose—and taking up in a pipette a measured volume of this, corresponding with, ay, 2 to 2½ times the volume of the undiluted blood which was employed, introduce his into the tube which contains the washed and drained clot. Now seal the upper

from the blood vessels, is overpowered. When this condition is arrive at and the pus fluid has become definitely tryptic, the trabeculæ c connective tissue between the tissue spaces, together with any strand of fibrin which may envelop the leucocytes, are rapidly dissolved, wit the result that an abscess sac is formed and definite fluctuation is obtained The pus fluid may now proceed to eat into the surrounding tissues, an then either burrow in the depth or point upon the surface.

Of these facts those which are material in connexion with the surviva of the infecting microbes are the following:—The fluid in the absces sac, being more or less completely shut off from the circulating lymp by a barrier of infiltrated tissue, furnishes a nidus for the growth of bac teria. Under the influence of those bacteria, the lymph in the nidus, whicl originally possessed both antibacterial and antitryptic properties, become first impoverished in antibacterial elements, then charged with bacteria products and finally (in particular where pyogenic micro-organisms, a distinguished from tubercle bacilli, are at work) endowed with trypti properties. It will be clear, *a priori*, on consideration of these facts, tha fresh phagocytes (should such arrive upon the scene) would, even, if they escaped paralysis by bacterial toxins and digestion by the tryptic ferment find in the abscess fluid no opsonic substances to co-operate with them.

The sequence of events which has just been described can, as a matte of fact, be readily followed by direct examination. While in the early stages which precede abscess formation phagocytosis may be encountered microscopic examination of pus after an abscess has been formed wil practically always show that the leucocytes have degenerated, and that phagocytosis is entirely absent. And separate examination of the

end of this tube in the flame and place in an incubator at a temperature of 50 to 55 C., which is the optimum temperature for a tryptic digestion.

Prepare, using in each case exactly the same volumes of blood, distilled water and gelatine, a whole series of tubes. Into certain of these introduce, mixing carefully with the gelatine, a measured volume of normal serum corresponding with $\frac{1}{4}$ to $\frac{1}{2}$ the volume of the blood originally employed.

After incubating the tubes at 50 to 55 C. for 2 to 3 days—that interval of time is required to allow of the disintegration of the leucocytes—examine the condition of the clot in the tubes, and place these upright in cold water or on the laboratory bench so as to give opportunity for the solidification of the gelatine. It will now be found that the clot will have disintegrated and the gelatine will have lost its power of setting in the case of the tubes which contain only gelatine and clot, while in the tubes which received an addition of serum, the tryptic action will have been inhibited, with the result that the clot will be intact and the gelatine will have retained its power of setting.

The measurement of the tryptic or antitrpytic power of a fluid may be carried out by the aid of throttled pipettes and the technique for progressive dilution, mixture and storage which I have described in connexion with the measurement of the agglutinating power of the blood (*Lancet*, July 25, 1903).

Into companion throttled pipettes there would be introduced (combined in the one case with serum and in the other case with an equivalent of water) progressive dilutions of the tryptic fluid mixed in each case with indicator fluid. That indicator fluid would consist of a milky suspension of very fine floccules of coagulated albumen, such as is obtained by heating a normal serum diluted sixfold with a very attenuated acetic acid. The clarification or, as the case may be, non-clarification of such a fluid by the progressive dilutions of the digestive fluid renders manifest to the eye the action of the digestive ferment or the inhibition of its action.

ucocytes and the pus fluid shows, in the case of the leucocytes, that, ven after they have been washed and have been furnished with normal erum, they are incompetent to ingest bacteria, and in the case of the us fluid that it paralyses healthy leucocytes, and that it is incapable f exerting any opsonic action upon bacteria which are obtained from e abscess.

(d) *Conditions which obtain where in connexion with a Microbic Infecon of the Skin a Nidus has been formed under the Shelter of a Scab.*—A ord or two will serve to make clear the conditions which obtain where surface infection has led to the formation of a scab. It will be appreiated that the outflow of lymph from the superficial vessels, which is ssociated with inflammatory response to a superficial infection, staunches s soon as the lymph begins to coagulate on the surface, and is definitely rrested when the coagulated lymph desiccates and hardens into a scab. In ssociation with this, such phagocytes as may be contained in that lymph vill be, in the first instance, immobilised, and then killed by desiccation. t will be quite otherwise with any surviving bacteria. These, when they ecome involved in the scab, will be sheltered from the attacks of the hagocytes. If now they succeed in multiplying in the deeper and corespondingly moister layers of the scab, they will reduce the antibacterial otency of the comparatively stagnant lymph which underlies the scab nd will then be in a position to invade the underlying subcutaneous issue or epithelium, and thus to extend the area of infection.

(e) *Conditions which obtain in the Case where we have to deal with a ersistent Sinus.*—Where a sinus is freely discharging pus the conditions re, I take it, not dissimilar from those which prevail in an abscess. The us, in such a case, possesses a low opsonic power, it contains no effective eucocytes, it may be charged with bacterial toxin, and it contains—as a lance at the sodden and digested appearance of the skin-surface in the eighbourhood of a discharging sinus informs us—a tryptic ferment.

In a case of a sinus which does not furnish any discharge we have, I take it, to deal with conditions somewhat comparable with those which would obtain in a well in the case where the inflowing water had deposited on its walls an insoluble element in such a manner as to choke all the conduits of inflow. Upon the walls and floor of such a well forms of life might quite well maintain themselves, which would be incapable of holding their own in the face of a copious inflow of water. So is it, I take it, in the case of the dry sinus. Here we may assume that the density of the granulation tissue which lines the walls, and the continual deposition of fibrin upon the surface of that lining membrane, prevent the free outflow of lymph on to the surface.

Consideration of the therapeutic measures by which we may bring the antibacterial agencies of the circulating blood into effective operation upon the microbes in a nidus of infection.

We have already appreciated that the spontaneous cure of localised

bacterial infections is effected by the transfer of antibacterial agencie from the circulating blood to the invaded tissues, and we have furthe appreciated that we can, in those cases of localised infection where spontaneous cure is not effected, give aid, on the one hand, by increasin the antibacterial resources which are at disposal in the circulating blood and, on the other hand, by bringing those antibacterial agencies int effective operation upon the bacteria in the invaded tissues. We hav here to consider the problem as to how the latter object can be achieve in the case where the invading bacteria have already made for themselve a nidus in the invaded tissues. This will involve the discussion of (1) *th measures which may be employed to favour the egress of antibacteria agencies from the blood,* and (2) *the measures which may be employed to ope the way for the entrance of the antibacterial substances and phagocytes int the actual nidus of infection.*

(1) *Measures which may be employed to favour the egress o Antibacterial Agencies from the Blood.* Although it might at first sight seem that it would be necessary, in connexion with the egress of antibacterial agencies from the blood to the tissues, to conside separately the case of the emigrating leucocytes and the transudin lymph, it becomes clear, on reflection, that, inasmuch as increased transudation is normally associated with increased emigration, and, inasmuch as we cannot promote the former of these without at the same time favouring the latter, it will be both legitimate and convenient to confine ourselves here to the discussion of the means we have at disposal for increasing transudation.

There are, in point of fact, two main factors which come into consideration in connexion with lymph—the first is the hydrostatic pressure in the local capillaries, the second is the coagulability and viscidity of the blood. I pass, therefore, to discuss the therapeutic measures we have at command for raising the hydrostatic pressure in the capillaries, and for diminishing the coagulability and viscidity of the blood.

(*a*) *Measures for raising the Hydrostatic Pressure in the Capillaries of the infected Part.*—It will be clear that, where the focus of infection is so situated as to make it possible to exercise control over the blood which circulates in it, there will be a possibility of increasing the hydrostatic pressure in the capillaries of the infected region, on the one hand, by determining a fuller supply of arterial blood to the affected region, and, on the other hand, by banking up the blood in the veins which carry off the blood from the affected part. We can raise the capillary pressure by the former method by applying hot fomentations, or heat in any other form, or rubefacients. We can raise the pressure by the latter method if, following the procedure of Bier, we apply a bandage loosely round the limb.

The consideration of the question as to how we can raise the hydrostatic pressure in the capillaries naturally leads on to the consideration of the question as to when and in what circumstances it will be expedient

o have recourse to measures for raising that pressure. Two different onsiderations must here be kept in view. On the one hand, the question s to whether the normal blood-supply to the infected part is capable f furnishing an adequate lymph-stream must be considered; on the other and, we must weigh the advantage which would be derived from the letermination of increased protective substances to the affected part gainst the disadvantages which might result from the fact that the ampler ymph-stream would carry into the blood bacterial products which vere previously locked up in the focus of infection. The decision as to vhere the balance of advantage lies in a particular case may thus be a ery delicate matter. It is in many cases a question which cannot be esolved without a trial supplemented by a series of blood-examinations. It may, however, be laid down as a general working rule that the advantage will be greatest where the bacterial focus is situated in a poorly vascularised tissue, and that the disadvantage will be least (*a*) in the earlier stages of an infection (*b*) where the bacterial focus is of strictly moderate dimensions, and (*c*) where it has just been evacuated. In the case where the dimensions of a bacterial focus are considerable, the disadvantages of determining an increased lymph-stream to the affected tissues will generally altogether outweigh the advantages.

(*b*) *Measures for diminishing the Coagulability and Viscidity of the Blood.*—In the course of my studies in connexion with the causation of urticaria and other forms of serous hæmorrhage, on the one hand, and thrombosis, on the other hand, I have repeatedly pointed out that a condition of diminished blood-coagulability is associated with increased transudation into the tissues, and that a condition of increased blood-coagulability is associated with restricted transudation and with the transudation of a lymph which is very prone to coagulate in the tissues. I have, further, in connexion with the studies here referred to, shown that transudation can be increased or diminished at pleasure by rendering the blood more coagulable by the exhibition of calcium salts or, as the case may be, by diminishing its coagulability by the administration of a decalcifying agent, such as citric acid (citric acid, 2–4 grammes, t.i.d.).

The fact that diminished coagulability of the blood goes hand in hand with increased transudation of lymph is of interest to us here, inasmuch as it suggests that diminished blood-coagulability may be a factor in the production of serous effusion.

The fact that increased blood-coagulability goes hand in hand with diminished transudation of lymph and with a tendency on the part of the effused lymph to clot in the meshes of the tissues, in like manner suggests that a hypercoagulable condition of the blood may be a factor in the production of brawny swelling and carbuncle.

The fact that transudation can be decreased by the administration of calcium salts has a possible therapeutic application in connexion with those cases where spontaneous auto-inoculations are occurring in re-

sponse to a conveyance of bacterial products into the blood by the agency of the lymph.

Lastly, the fact that lymph transudation may be increased, and the fact that the tendency on the part of a lymph to coagulate in the tissues may be counteracted by the administration of citric acid, has, it seems to me, a very important application in connexion with the treatment of brawny swelling. I have already [1] given details of a case of Ludwig's angina, where an obvious change for better ensued upon the administration of this decalcifying and lymphagogic agent.

In this connexion it is interesting to note that our grandmothers employed with success a similar method of treatment in connexion with infections of the respiratory passages. They were, as you remember, wont to administer decalcifying draughts of black currant tea for the purpose of "loosening a hard cough," and they often followed these up by another lymphagogue in the form of oatmeal gruel.

(2) *Measures that may be employed to open the way for the entrance of Antibacterial Elements into the Focus of Infection.*—We have already, in connexion with the suggestion that citric acid may with advantage be administered for the purpose of counteracting the clotting of the lymph in the focus of infection, considered one of the measures that may be employed to facilitate the entrance of phagocytes and antibacterial elements into the focus of infection. There are, however, other obstacles to that entrance. There is, in the first place, the barrier which is opposed by the infiltration of the tissues by leucocytes, and there is again the obstacle which is constituted by the accumulation of fluid in the focus of infection. Moreover, as we have seen when we were considering the conditions which obtain in abscesses, there are other hindrances, in the form of bacterial toxins and digestive ferments, which interfere with the functioning of the phagocytes. All these obstacles and hindrances can be got out of the way only by resort to evacuation.

We may briefly consider four different methods of evacuation—(*a*) *evacuation by incision*; (*b*) *evacuation by aspiration*; (*c*) *evacuation by puncture, or incision. combined with cupping*; (*d*) *evacuation by puncture, or incision or removal of scabs combined with the local application of a chemical lymphagogue.*

(*a*) *Evacuation by Incision.*—It is an axiom in surgery that all pus must be cut down upon, and it is almost equally a precept of surgical obligation, to carry free incisions through the infiltrated tissues where an inflammatory reaction culminates, instead of in pus, in brawny swelling, or in carbuncle. It is perfectly well understood in the case of abscess—not so well understood, I think, in connexion with brawny swelling or carbuncle—that the object of incision is in all cases the evacuation of the products of inflammation. We have witness to this in the fact that, in the case of suppurative processes, operative interference is postponed until fluctuation is obtained, and that provision is made for drainage either by opening

[1] *Lancet*, August 24, 1907.

ne abscess at the most dependent part or by introducing tubes into the ound. But where, in the case of brawny swelling, incision has not resulted n evacuation, the surgeon does not seek a remedy for this condition. And he illogicality of surgery does not stop here. While in certain cases the vacuation of the contents of an abscess is followed up by measures which ring phagocytes and antibacterial substances from the blood into the vacuated focus—I am thinking here of hot fomentations—in most cases vacuation is regarded as if it were an end in itself, instead of being only means for bringing the antibacterial agencies of the blood into effective peration. And yet where this is not achieved by incision, nothing is ccomplished in the matter of the extinction of the infection. In fact, in uch a case, nothing useful is achieved by operation, unless it be the afeguarding of the skin from erosion by the tryptic ferment of the pus, he prevention of burrowing, and the arrest of absorption.

(b) *Evacuation by Aspiration.*—Between evacuation by incision and vacuation by aspiration there are only few and comparatively unimport-nt differences. From the point of view of the complete evacuation of he contents of the abscess the advantage clearly rests with the method f incision. From the point of view of the avoidance of an external ound with the opportunity it furnishes for secondary infection, the dvantage equally clearly lies with the method of aspiration. There emains only the question of the greater or less freedom of access of anti-acterial agencies to the nidus of infection in either case, and the ques-ion of drainage. On the whole the advantage would here seem to rest ith the method of aspiration, for no sooner is an abscess sac evacuated han it begins to fill up again with an antitryptic and opsonic lymph, and he equivalent of drainage is easily provided by repeating the aspiration.

(c) *Evacuation by Incision or Multiple Puncture combined with Cup-ing.*—The procedure here in question is a procedure which is recom-nended by Bier and Klapp as useful in connexion with the treatment f furuncles and carbuncles and localised tubercular affections.

At first sight, this method would seem to furnish exactly the kind of notive force which an immunisator would wish to employ for evacuating he inspissated inflammatory products from the choked tissues, and for rawing out from the blood, and carrying through the infiltrated or hyper-lasic tissues, a stream of immunising lymph.

Practical experience and reflection, however, make clear that one equisite to success has here been overlooked.

In reality the procedure of Bier and Klapp is a filtration process arried out with the help of an exhaust. And it will be clear that where viscid and coagulable fluid has to be drawn through a very fine system f pores, the clotting of the fluid in or at the mouths of those pores may ery readily bring the filtration to a standstill. In the case of a laboratory xperiment, the filter, if it is a paper filter, will then bulge and give way. n the case of the animal tissues in like manner, even when the negative ressure is kept within strict limits, inevitably something will give way.

What will in point of fact give way will be the delicate capillary wall and in the case of a carbuncle thus treated its last state is, in my exper ence, worse than the first.

(*d*) *Evacuation by Incision, Puncture, or Removal of Scabs combine with the local Application of a chemical Lymphagogue.*—Attention has ju been directed to the defects which must attach to every method whic seeks to achieve by mechanical force the evacuation of obtruding materi and the passage of a coagulable fluid through a choked filter.

There is a more excellent way.

If we can either clear the surface of the filter (I am here thinking the removal of scabs from an infected surface), or pierce through th surface layer of the filter (I am thinking here of incision or puncture int a nidus of infection), and if we can then, without the application of an violence, cause the fluid to well out through the pores, depriving the same time that fluid of its coagulability, we shall gradually open u and then keep open our choked filter.

I have already, on more than one occasion, pointed out that we hav in a citrated hypertonic salt solution a chemical agent which will, applie to the surface of any open system of tissue spaces, cause a lymph strea to set outwards through those tissue spaces towards the surface, whi it will, at the same time, deprive that lymph of its coagulability.

I have suggested that this agent should be prescribed in the form a powder consisting of five parts of common salt and one half part citrate of soda, with instructions that it should be dissolved whe required in 100 parts of boiling water. A concentration of 1·5 to 2 pe cent. of salt will, however, in certain cases be preferable, the concentra tion of the citrate remaining as before at 0·5 per cent.

Where incisions have been carried down into infiltrated tissues, li soaked in the stronger solution may with advantage be brought int the wounds.

In the case where an abscess cavity cannot be effectively draine the stronger solution may with advantage be introduced into the sac, th fluid which collects being of course, from time to time, evacuated. Wher as in the thick pus obtained before treatment, phagocytosis will have bee absent, it will, if the blood is not gravely at fault, be conspicuous in th thin sero-pus which now collects in the sac.

Sinuses may with advantage be syringed out with the same lotio a piece of lint soaked in the lotion being afterwards introduced into the orifices. Here again, after the citrate and salt application, evidenc of phagocytic activity will be found on examining the discharge. If no the fault may with advantage be sought in the blood.

Keeping in view the fact that it is always possible, by continuousl "drawing," to keep an orifice permanently open, it will be advisable the case where we are dealing with a sinus, to suspend the citrate an salt applications from time to time, for a few days at a time, to ascerta what has been accomplished.

In the case of a boil or a tubercular abscess, it will be well, after the ıs has been evacuated, to apply the citrate and salt solution to the orifice, ıd to continue to do so as long as any induration persists or any definitely ırulent discharge is obtained.

The skin in the neighbourhood of the orifice may with advantage be otected against the irritating effect of the brine by a coating of vaseline.

In the case where a surface is covered with scab, it will be advisable, ter this has been removed, to apply the weaker solutions of citrate and lt, and to continue the applications only for a few minutes to half an our.

After the wound has in this way been sufficiently washed with a stream immunising lymph, it will be well to staunch the lymph flow by powering the surface with a styptic powder, consisting of calcium chloride part, precipitated chalk 400 parts.

It is, I think, of interest, in connexion with these therapeutical appliations of the citrate and salt, to recall that we have in this decalcifyıg and hydragogue application only a variant of the soap and sugar lasters which our grandmothers were wont to employ for "drawing" oils.

A similar interest attaches to the fact that another of the therapeutic pplications of this citrate and salt solution was anticipated by the West ndian slave-owners.

I find it recorded of them that it was their practice, when they had courged a slave and had cut the skin of his back into ribbons, to prevent ne supervention of gangrene by rubbing into the wounds a concentrated rine, fortified by a decalcifying agent in the form of the juice of green emons, and a further lymphagogue in the form of allspice. It was rutal, but it was very intelligent.

Finally, in this connexion we may note that brine springs and sea vater have acquired, and probably have deserved, a certain reputation ı connexion with the treatment of scrofulous ulceration.

rganisation of the Medical Profession for carrying out the Work of Therapeutic Immunisation.

In conclusion, a final word may be appropriate on the subject of the rganisation of the medical profession for carrying out the work of theraeutic immunisation.

I have already said that, in the future of my expectation, the physician vill be an immunisator. I conceive that the task of making, by bacterioogical methods, a direct or inferential diagnosis of the nature of the atient's infection, the task of preparing and standardising special vaccines, the task of controlling the output of protective substances by lood examinations, and the direction of immunisation procedures vhere blood examinations are required as a guide will all fall upon his houlders. To carry out these difficult and delicate tasks the physician vill needs have to be trained as a laboratory worker.

But the physician—so I conceive of the matter—will not be the onl immunisator.

Upon the general practitioner, when he shall have been trained i the physiology of immunisation, as he is now trained in the physiolog of the circulatory system or the digestive system, will devolve, I take i all such therapeutic immunisation, whether it be in the form of vaccin therapy or auto-inoculation, as it shall prove practicable to reduce to system of routine, or to conduct under the control of the clinical sym toms.

And upon the general practitioner, or, as the case may be, upon the su geon, will devolve the task of directing such antibacterial agencies as ma be available in a patient's blood to the destruction of microbes in the loc focus of infection. When the surgeon comes to regard it as his functio in connexion with bacterial infections to minister to immunisation h will not, in the case where he has to deal with infiltrated and infecte tissues, stop short at mere incision and drainage, but will work to secu that free lymph flow through the focus of infection which is essential to im munisation. In like manner, when he has to deal with a suppurating focu he will not rest satisfied with evacuating the pent-up pus, but will recognis that such evacuation contributes to the extinction of the infection only i such measure as it serves to bring the antibacterial agencies of the bloo into effective operation upon the pathogenetic microbes. And again, whe the surgeon has to deal with a wound which refuses to heal, or with wound which is pouring out day by day a wholly ineffective pus, he wi realise that what is required in such a case is an increase of the ant bacterial power of the blood and a more effective lymph flow such a would bring the antibacterial agents of the blood into active operatio upon the infecting microbes.

Conclusion.

In conclusion, let me say this, that while I anticipate that the metho of increasing the antibacterial powers of the blood will be constantl improved, and while better methods for measuring the antibacteri power of the blood than any which we now have at disposal wi no doubt be discovered, and while indubitably more effective methods fo bringing the antibacterial agencies of the blood into operation in th focus of infection will be devised, we have already gone far enoug in the path of therapeutic immunisation to see that we have in the powe of increasing the antibacterial power of the blood by the agency of vaccine and in our power of bringing the antibacterial agencies of the bloo into operation in the focus of infection beyond all comparison the mos valuable assets in medicine.

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